FIG. 1

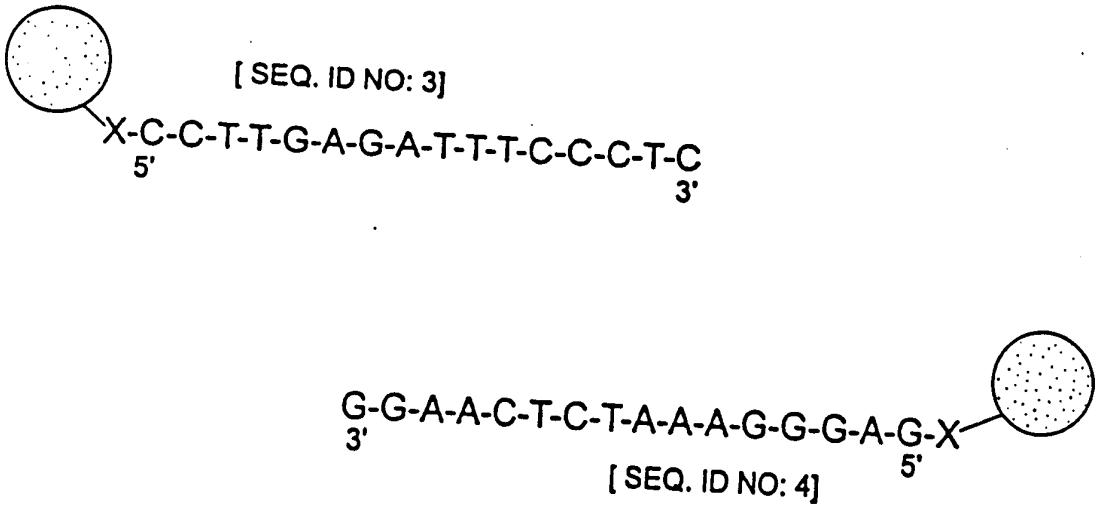

[SEQ. ID NO: 3]
X-C-C-T-T-G-A-G-A-T-T-T-C-C-C-T-C
5'
3'
G-G-A-A-C-T-C-T-A-A-A-G-G-G-A-G-X
3'
5'
[SEQ. ID NO: 4]
X-C-C-T-T-G-A-G-A-T-T-T-C-C-C-T-C
G-G-A-A-C-T-C-T-A-A-A-G-G-G-A-G-X

FIG.2

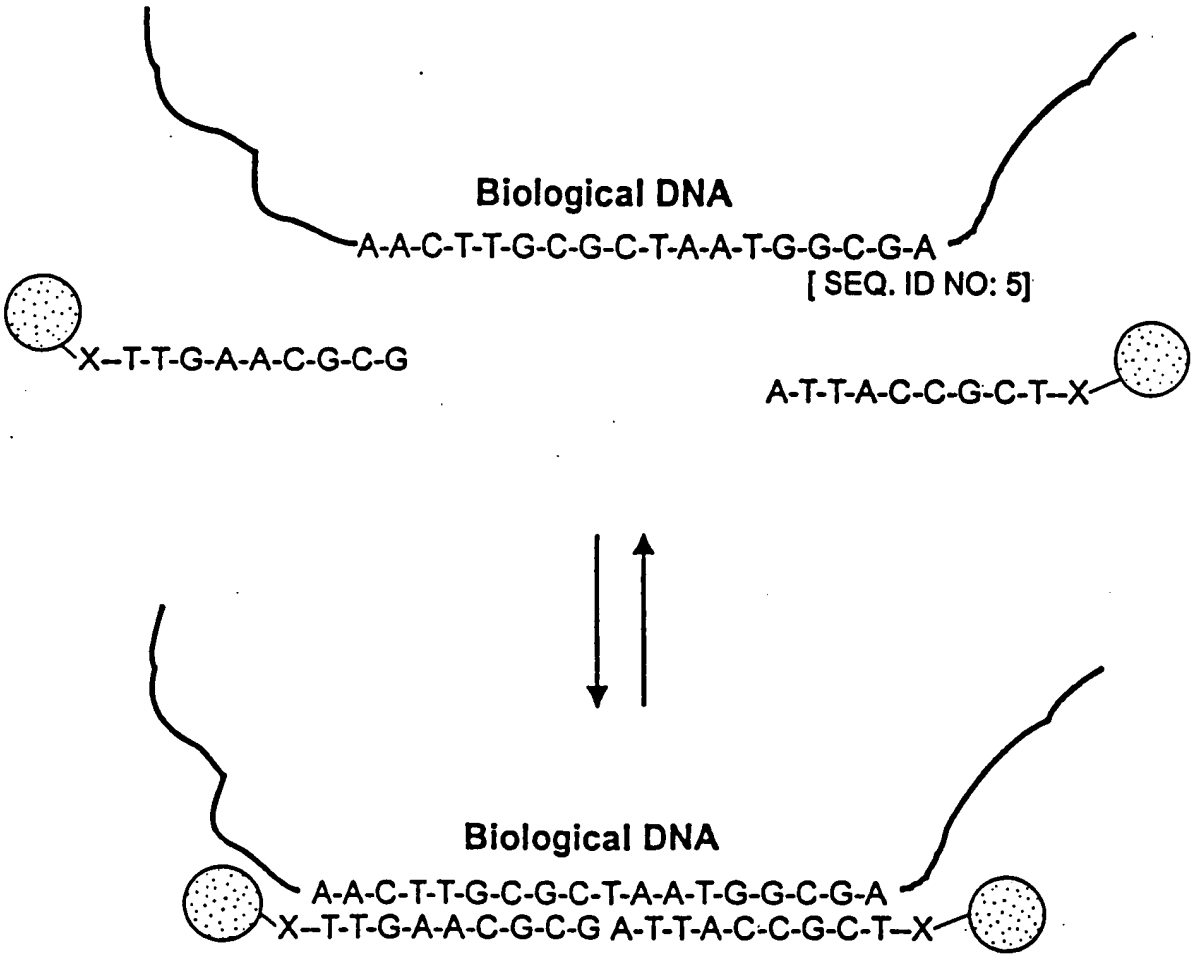

Biological DNA
A-A-C-T-T-G-C-G-C-T-A-A-T-G-G-C-G-A
[SEQ. ID NO: 5]
X–T-T-G-A-A-C-G-C-G
A-T-T-A-C-C-G-C-T–X
Biological DNA
A-A-C-T-T-G-C-G-C-T-A-A-T-G-G-C-G-A
X–T-T-G-A-A-C-G-C-G A-T-T-A-C-C-G-C-T–X

FIG.3

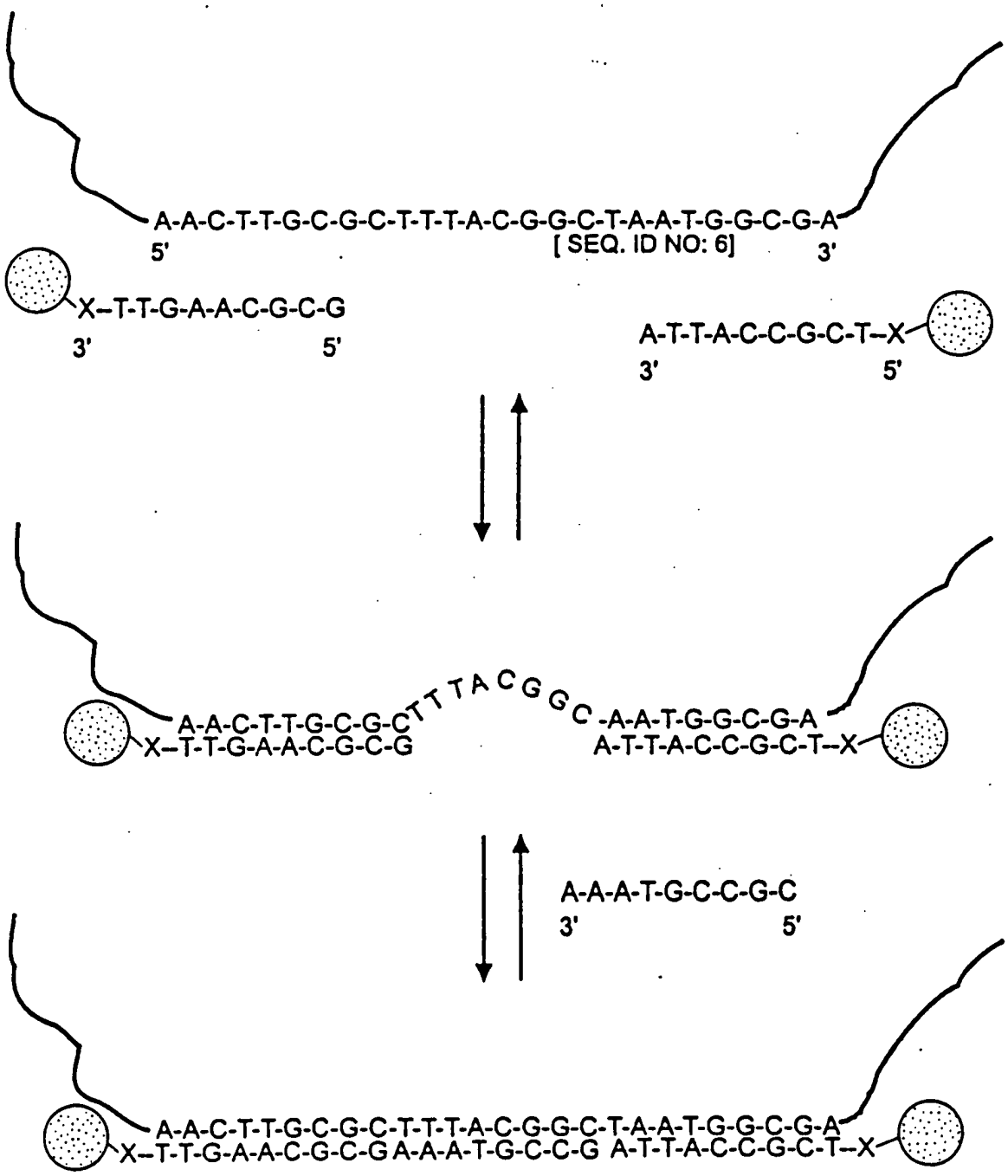

A-A-C-T-T-G-C-G-C-T-T-T-A-C-G-G-C-T-A-A-T-G-G-C-G-A
5'
[SEQ. ID NO: 6]
3'
X–T-T-G-A-A-C-G-C-G
3'
5'
A-T-T-A-C-C-G-C-T–X
3'
5'
A-A-C-T-T-G-C-G-C
T T T A C G G C
-A-A-T-G-G-C-G-A
X–T-T-G-A-A-C-G-C-G
A-T-T-A-C-C-G-C-T–X
A-A-A-T-G-C-C-G-C
3'
5'
A-A-C-T-T-G-C-G-C-T-T-T-A-C-G-G-C-T-A-A-T-G-G-C-G-A
X–T-T-G-A-A-C-G-C-G A-A-A-T-G-C-C-G A-T-T-A-C-C-G-C-T–X

FIG.4

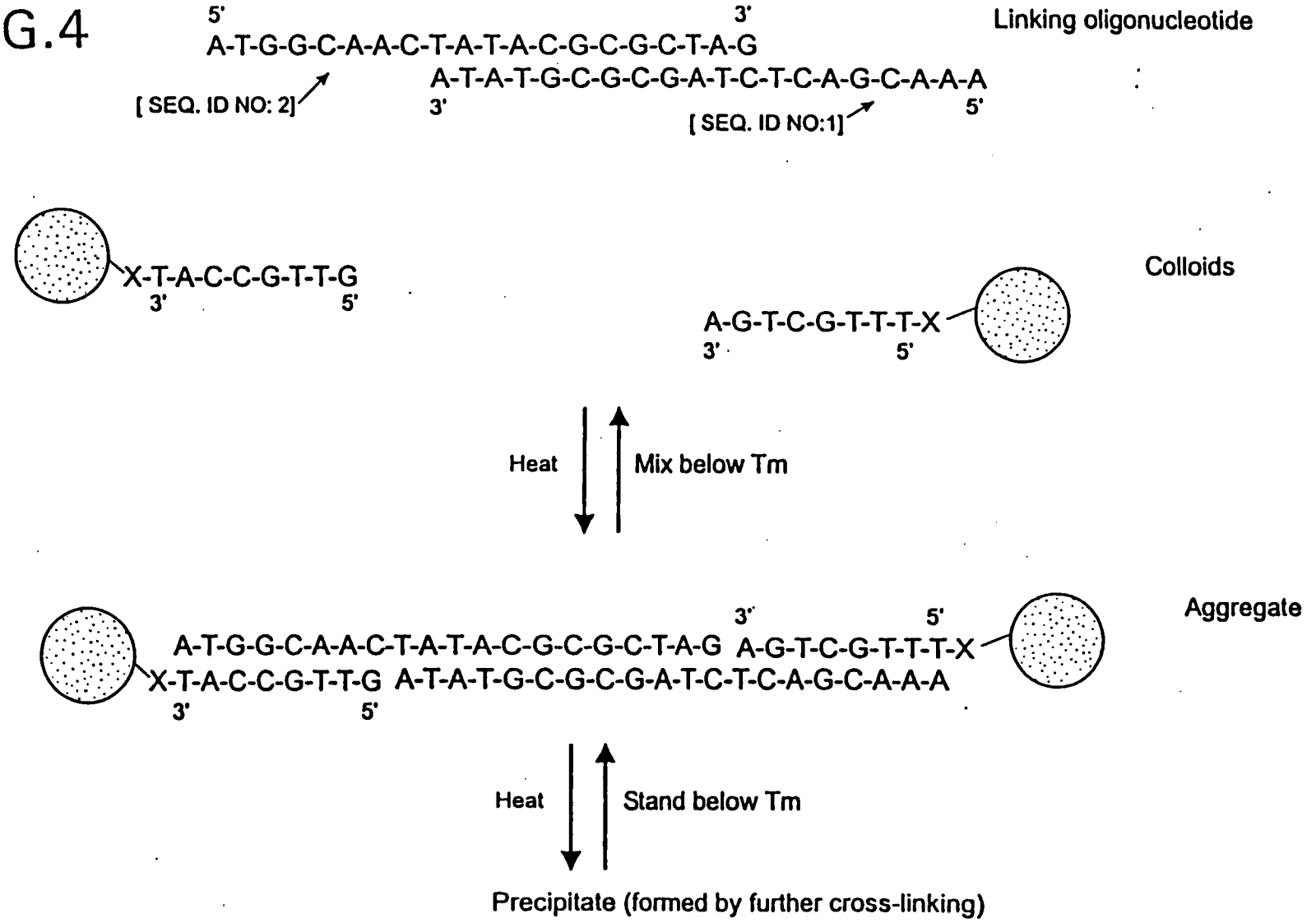
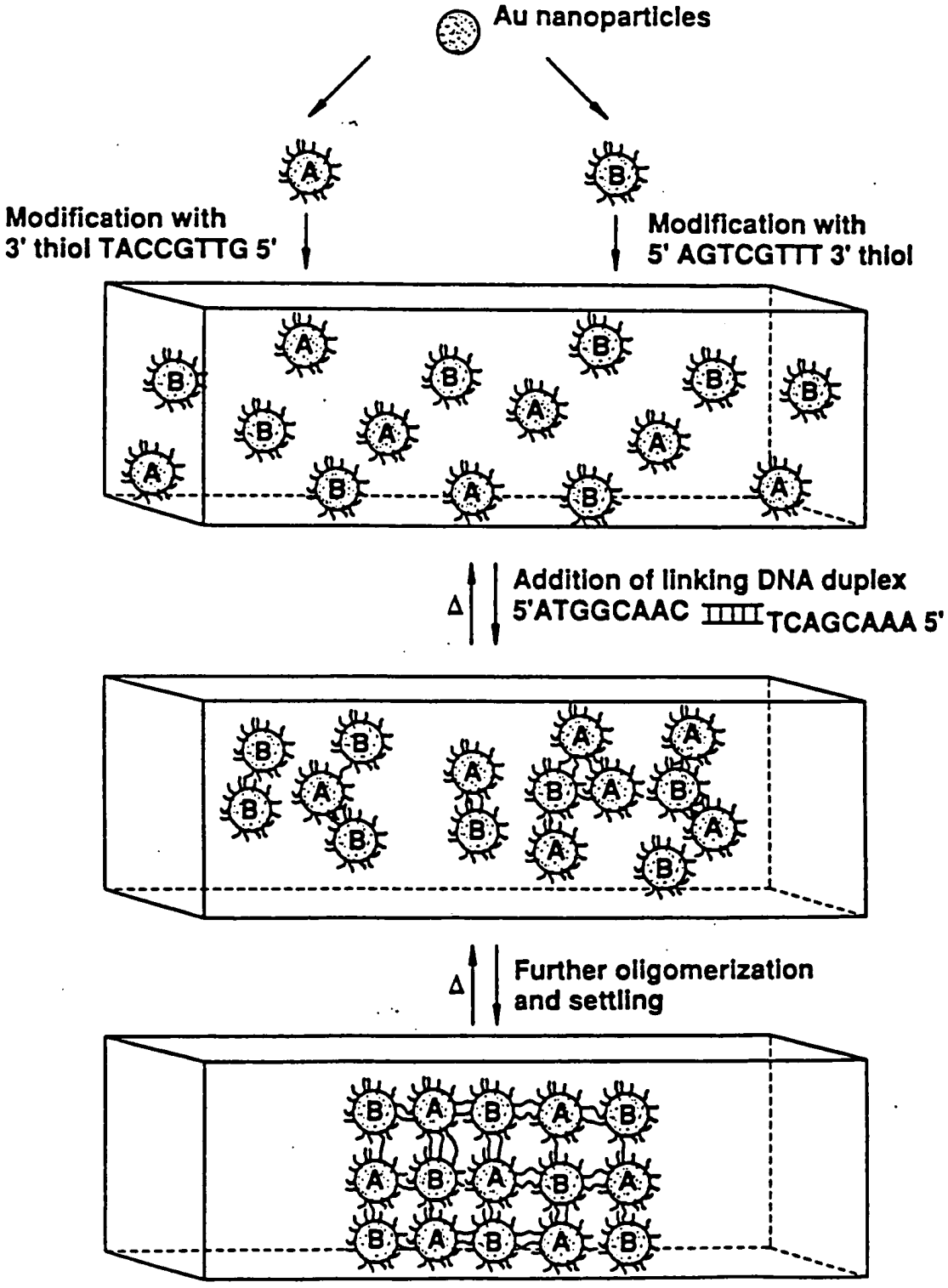

5'
3'
Linking oligonucleotide
A-T-G-G-C-A-A-C-T-A-T-A-C-G-C-G-C-T-A-G
A-T-A-T-G-C-G-C-G-A-T-C-T-C-A-G-C-A-A-A
[SEQ. ID NO: 2]
3'
5'
[SEQ. ID NO:1]
X-T-A-C-C-G-T-T-G
3'
5'
Colloids
A-G-T-C-G-T-T-T-X
3'
5'
Heat
Mix below Tm
3'
5'
Aggregate
A-T-G-G-C-A-A-C-T-A-T-A-C-G-C-G-C-T-A-G A-G-T-C-G-T-T-T-X
X-T-A-C-C-G-T-T-G A-T-A-T-G-C-G-C-G-A-T-C-T-C-A-G-C-A-A-A
3'
5'
Heat
Stand below Tm
Precipitate (formed by further cross-linking)

FIG.5


Au nanoparticles
A
B
Modification with
3' thiol TACCGTTG 5'
Modification with
5' AGTCGTTT 3' thiol
Δ
Addition of linking DNA duplex
5'ATGGCAAC
TCAGCAAA 5'
Δ
Further oligomerization
and settling

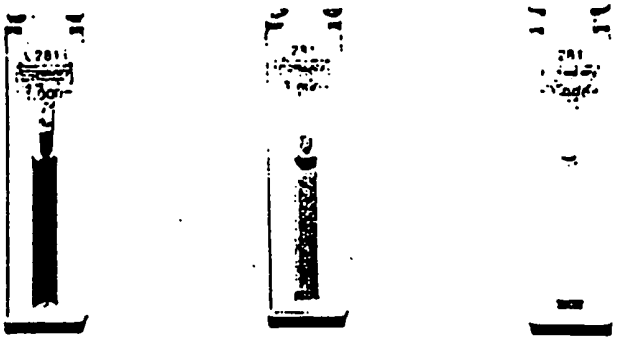


FIG.6A FIG.6B FIG.6C

FIG.7

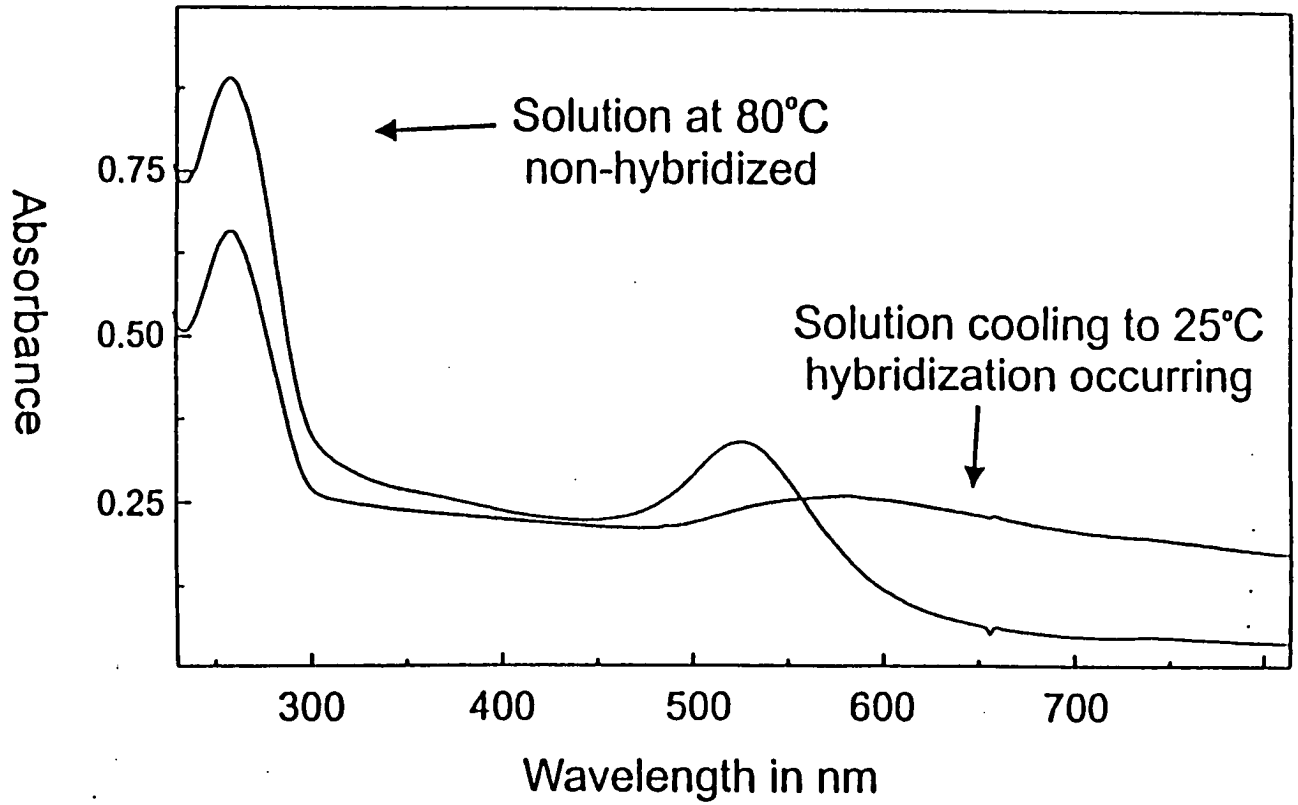


FIG.8A

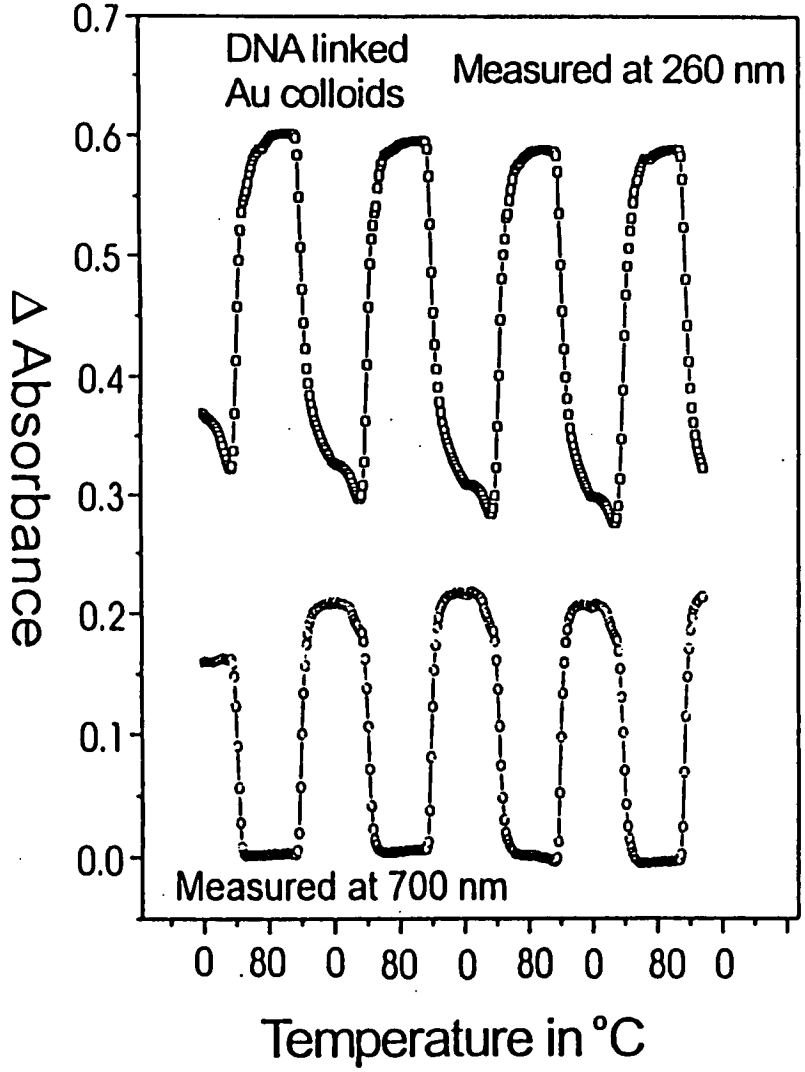

DNA linked
Au colloids
Measured at 260 nm
Measured at 700 nm
Δ Absorbance
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
0
80
0
80
0
80
0
80
0
Temperature in °C

FIG.8B

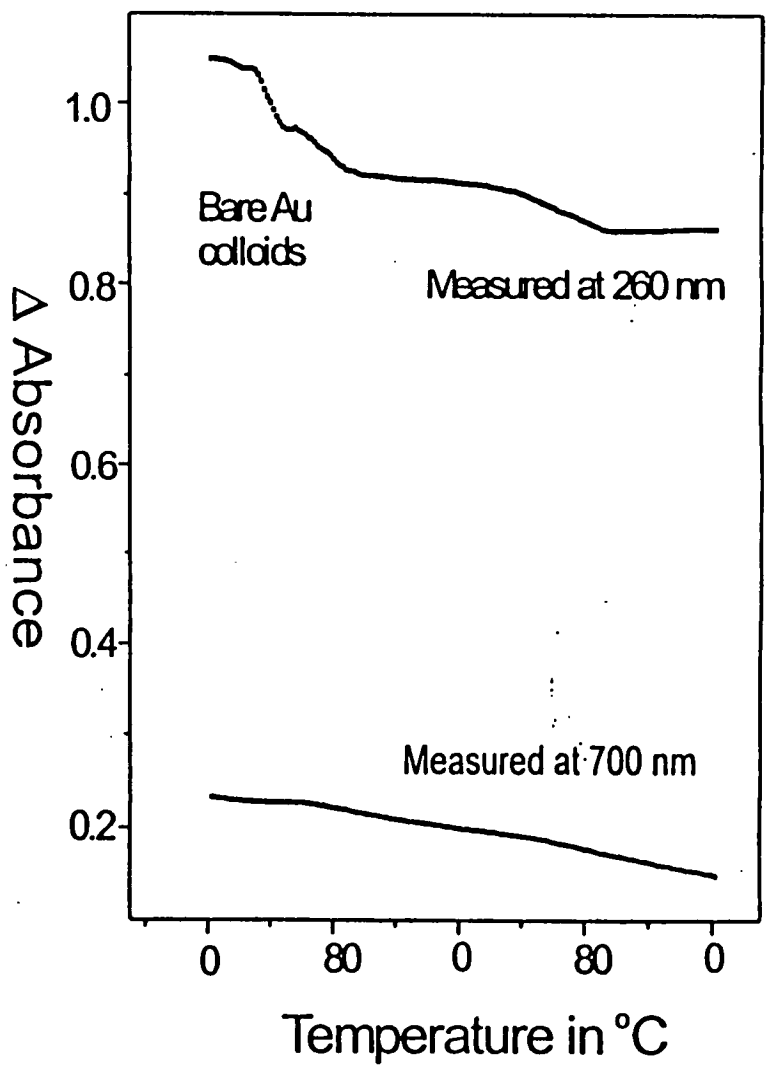

Bare Au
colloids
Measured at 260 nm
Measured at 700 nm
Δ Absorbance
1.0
0.8
0.6
0.4
0.2
0
80
0
80
0
Temperature in °C

FIG.9A

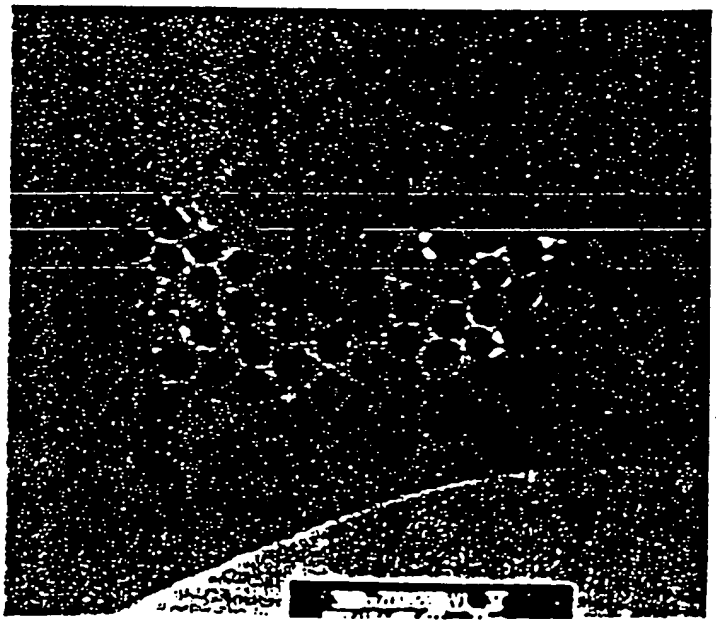


FIG.9B

FIG.10

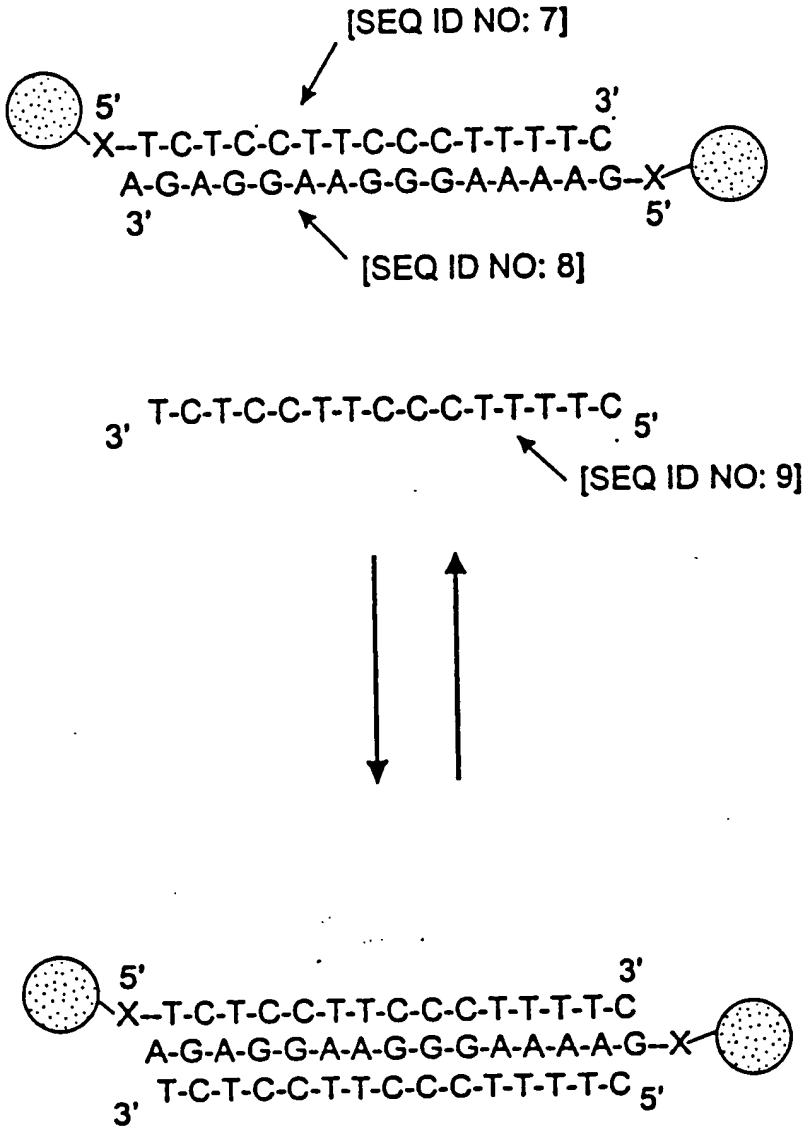

[SEQ ID NO: 7]
5'
3'
X--T-C-T-C-C-T-T-C-C-C-T-T-T-T-C
A-G-A-G-G-A-A-G-G-G-A-A-A-A-G--X
3'
5'
[SEQ ID NO: 8]
3'
T-C-T-C-C-T-T-C-C-C-T-T-T-T-C
5'
[SEQ ID NO: 9]
5'
3'
X--T-C-T-C-C-T-T-C-C-C-T-T-T-T-C
A-G-A-G-G-A-A-G-G-G-A-A-A-A-G--X
3'
T-C-T-C-C-T-T-C-C-C-T-T-T-T-C
5'

FIG. 11

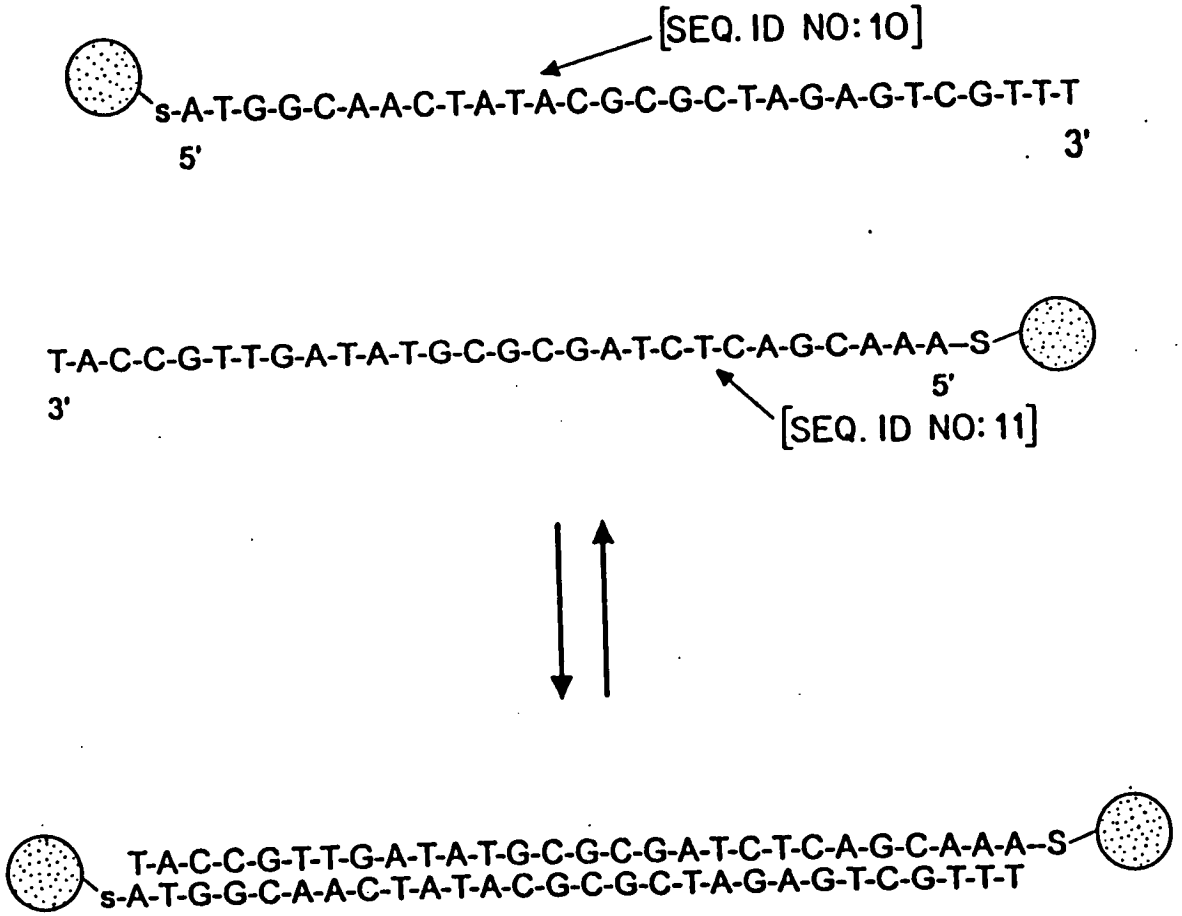

[SEQ. ID NO: 10]
s-A-T-G-G-C-A-A-C-T-A-T-A-C-G-C-G-C-T-A-G-A-G-T-C-G-T-T-T
5'
3'
T-A-C-C-G-T-T-G-A-T-A-T-G-C-G-C-G-A-T-C-T-C-A-G-C-A-A-A-S
3'
5'
[SEQ. ID NO: 11]
T-A-C-C-G-T-T-G-A-T-A-T-G-C-G-C-G-A-T-C-T-C-A-G-C-A-A-A-S
s-A-T-G-G-C-A-A-C-T-A-T-A-C-G-C-G-C-T-A-G-A-G-T-C-G-T-T-T

FIG.12A

Complementary Target

FIG.12B

Probes without Target

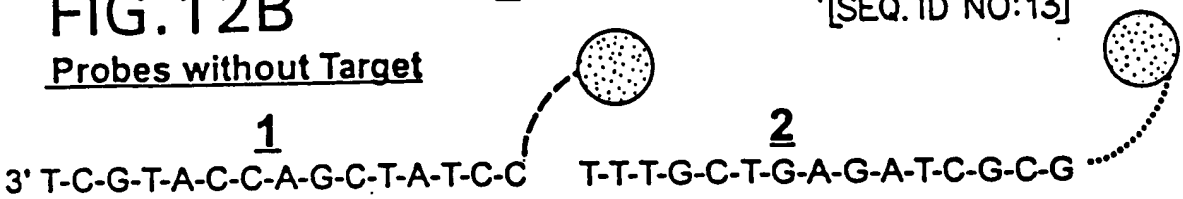


FIG.12C

Half Complementary Target

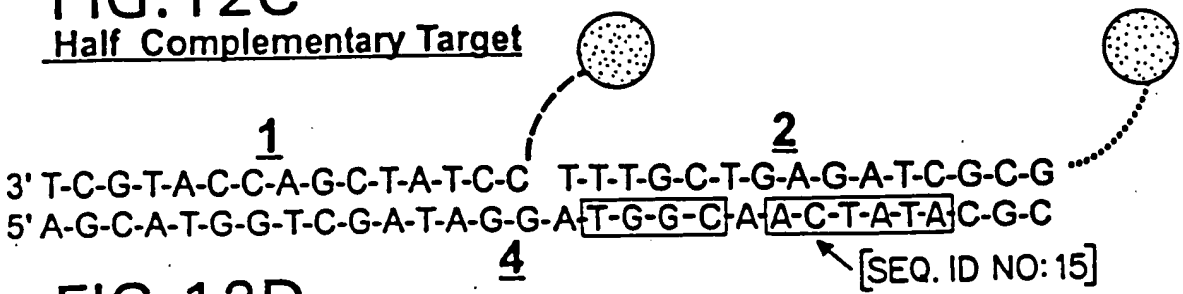


FIG.12D

Target - 6 bp

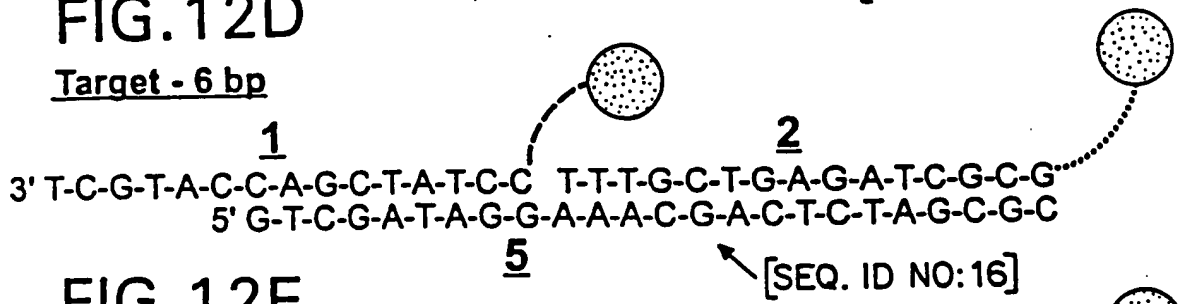


FIG.12E

One bp Mismatch

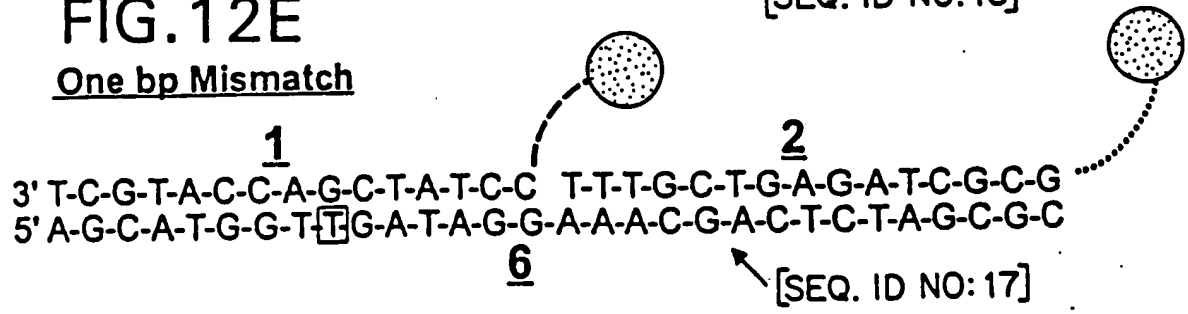


FIG.12F

Two bp Mismatch

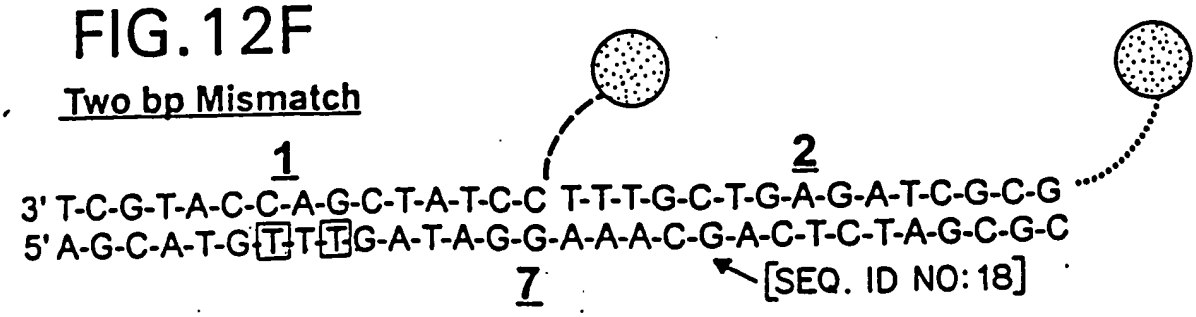
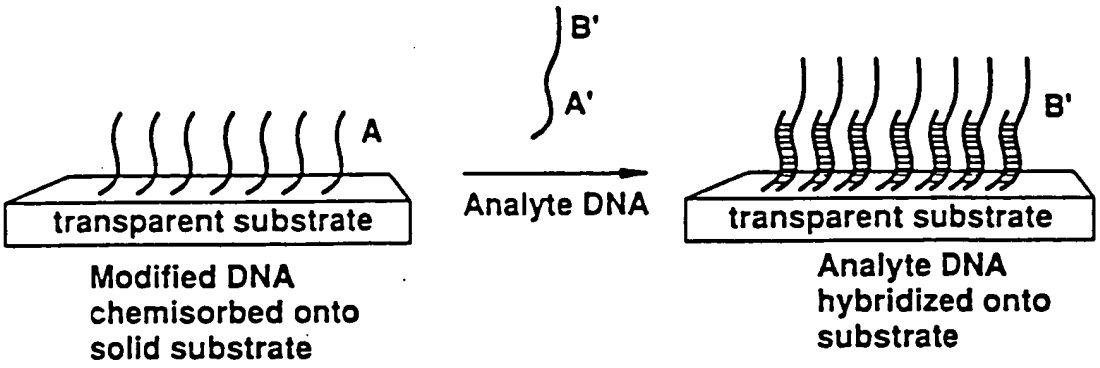
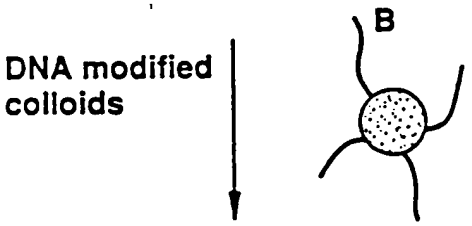


FIG.13A


B'
A'
A
transparent substrate
Analyte DNA
B'
transparent substrate
Modified DNA chemisorbed onto solid substrate
Analyte DNA hybridized onto substrate


DNA modified colloids
B

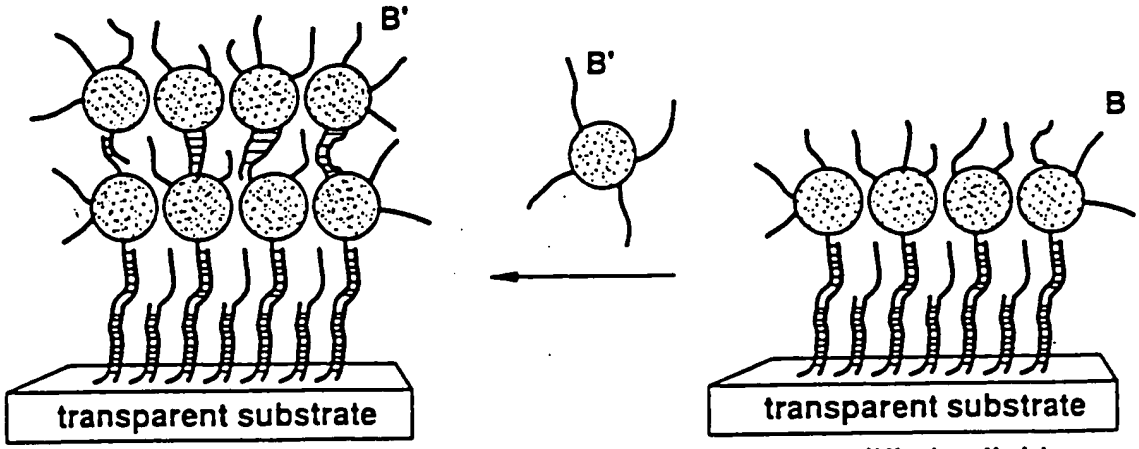

B'
B'
B
transparent substrate
transparent substrate
Dark areas where nanoparticle aggregates are linked to substrate surface by analyte DNA
DNA modified colloids hybridized to bound analyte DNA

FIG.13B

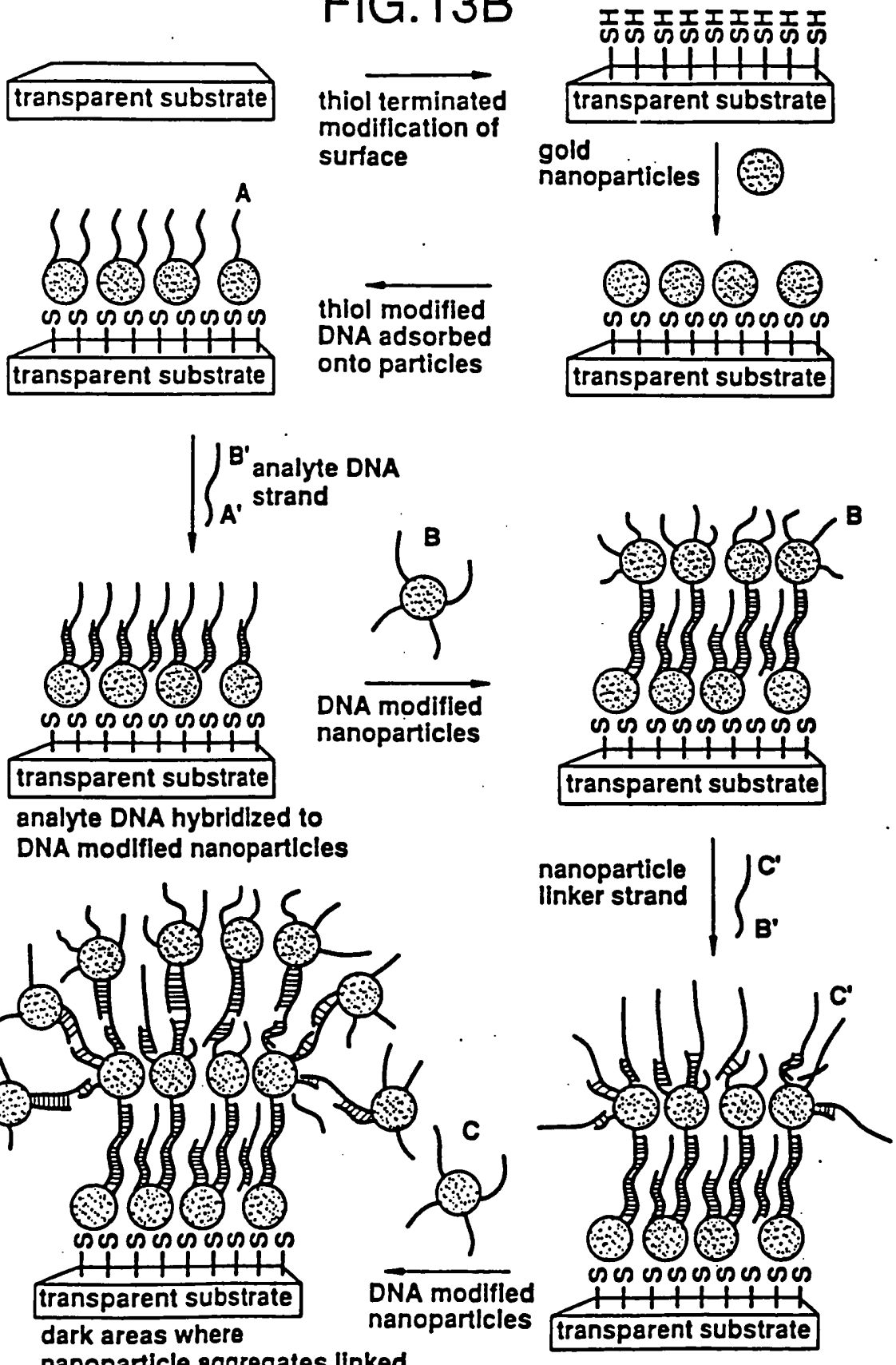

transparent substrate
thiol terminated modification of surface
transparent substrate
gold nanoparticles
A
thiol modified DNA adsorbed onto particles
transparent substrate
transparent substrate
B'
A'
analyte DNA strand
B
DNA modified nanoparticles
transparent substrate
B
transparent substrate
analyte DNA hybridized to DNA modified nanoparticles
nanoparticle linker strand
C'
B'
C
DNA modified nanoparticles
C'
transparent substrate
transparent substrate
dark areas where nanoparticle aggregates linked to substrate by analyte DNA

FIG.14A

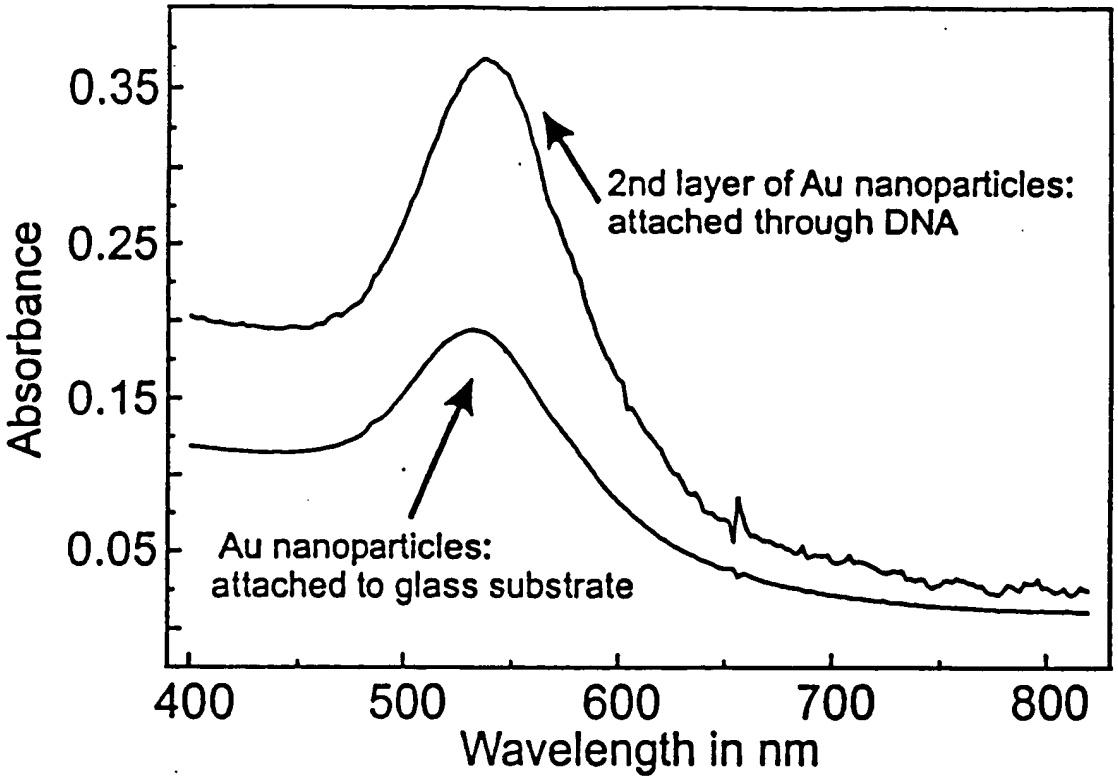


FIG.14B

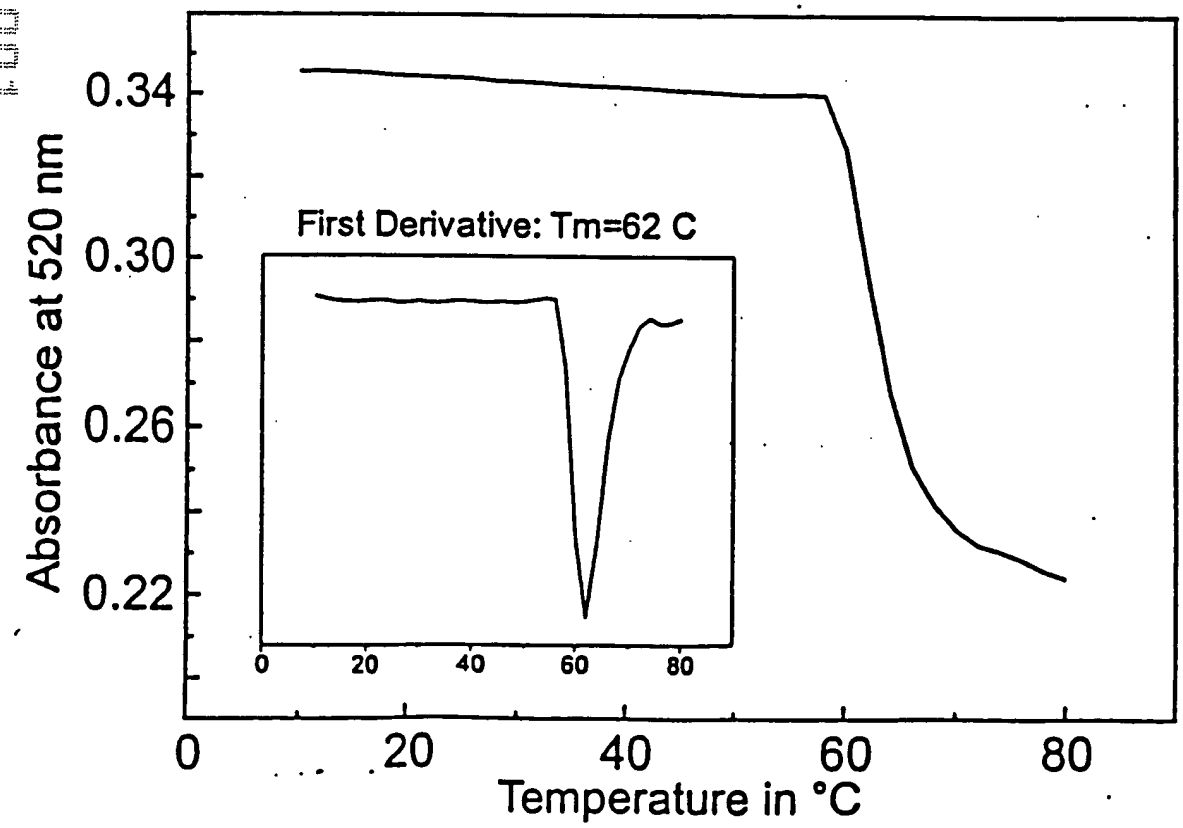


FIG15A

Probes with No Target

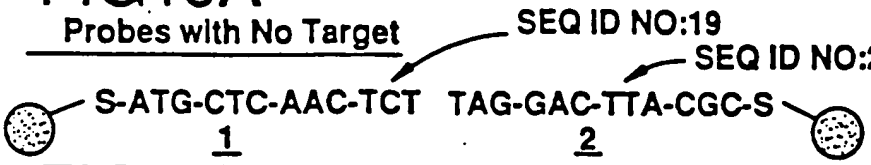


FIG15B

Half-Complementary Target

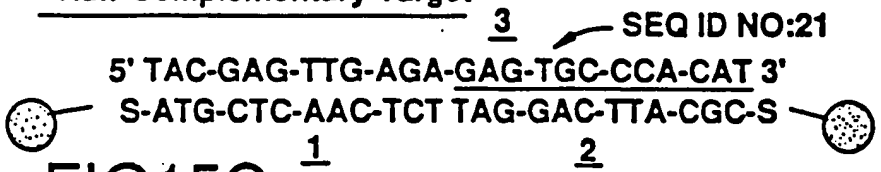


FIG15C

Complementary Target

Tm=53.5°C

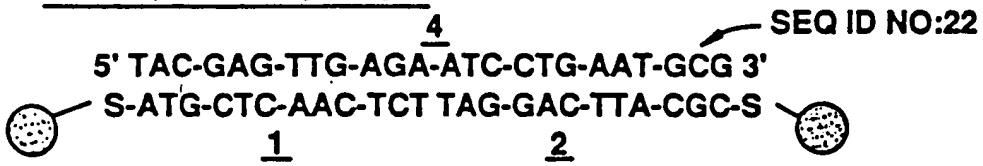


FIG15D

ONE Base-Pair Mismatch at Probe Head

Tm=50.4°C

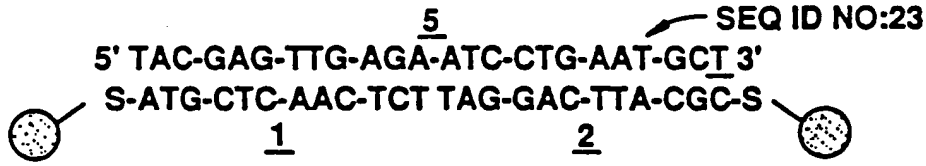


FIG15E

Tm=46.2°C

ONE Base-Pair Mismatch at Probe Tail

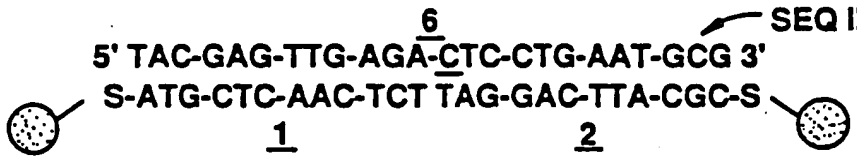


FIG15F

Tm=51.6°C

ONE Base Deletion

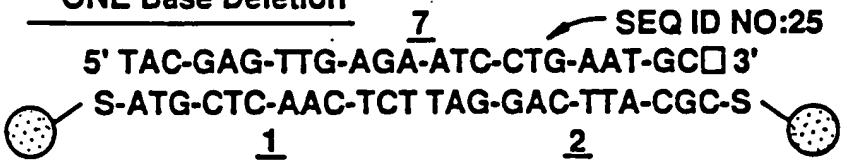


FIG15G

Tm=50.2°C

ONE Base-Pair Insertion

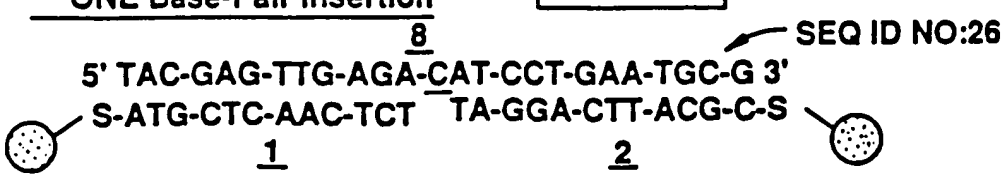


FIG.16A **24 Base Template**

5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GCG 3'
S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S
1 2

FIG.16B **48 Base Template with Complementary 24 Base Filler**

5' TAC-GAG-TTG-AGA-CCG-TTA-AGA-CGA-GGC-AAT-CAT-GCA-ATC-CTG-AAT-GCG 3'
S-ATG-CTC-AAC-TCT GGC-AAT-TCT-GCT-CCG-TTA-GTA-CGT TAG-GAC-TTA-CGC-S
1 2

FIG.16C **72 Base Template with Complementary 48 Base Filler**

5' TAC-GAG-TTG-AGA-CCG-TTA-AGA-CGA-GGC-AAT-CAT-GCA-TAT-ATT-GGA-CGC-TTT-ACG-GAC-AAC-ATC-CTG-AAT-GCG 3'
S-ATG-CTC-AAC-TCT GGC-AAT-TCT-GCT-CCG-TTA-GTA-CGT-ATA-TAA-CCT-GCG-AAA-TGC-CTG-TTG TAG-GAC-TTA-CGC-S
1 2

FIG.17A

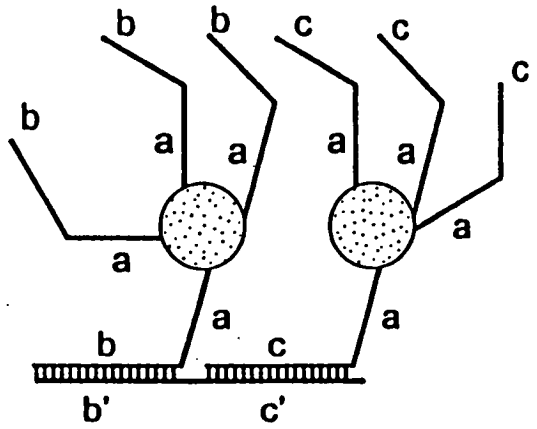


FIG.17B

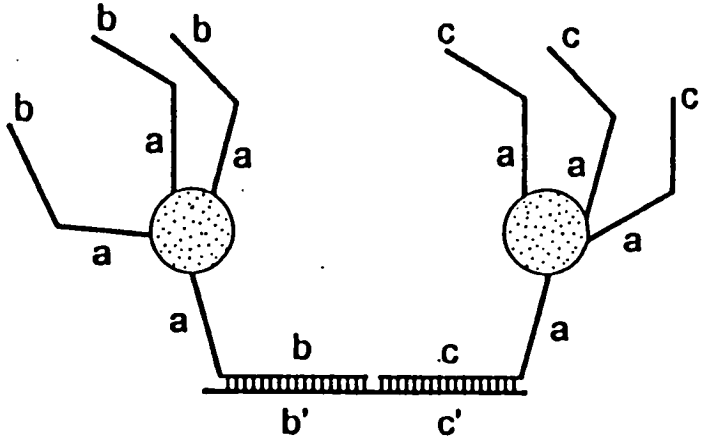


FIG.17C

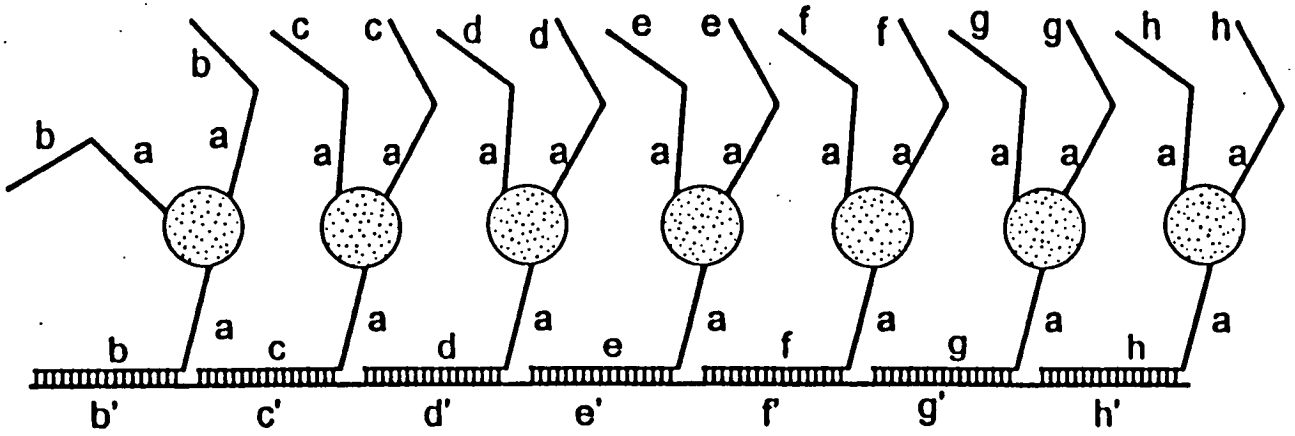


FIG.17D

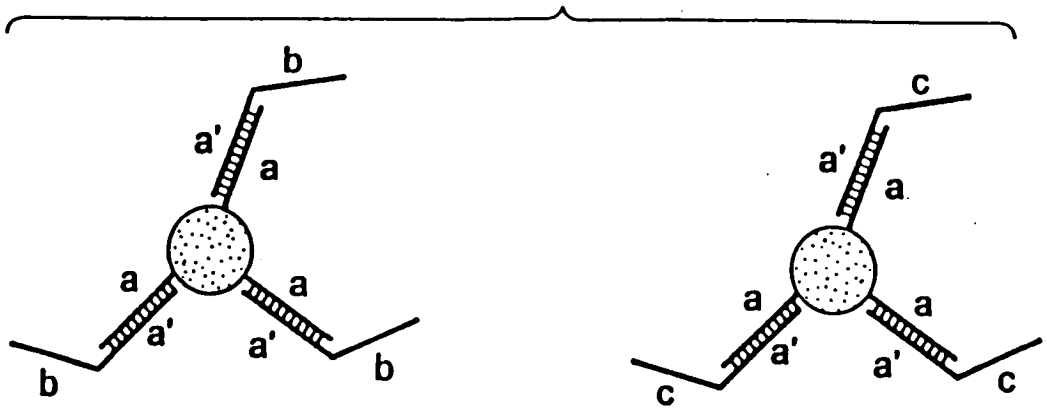


FIG.17E

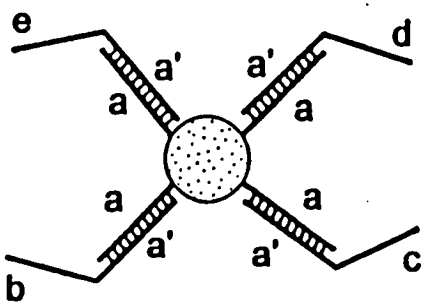


FIG.18

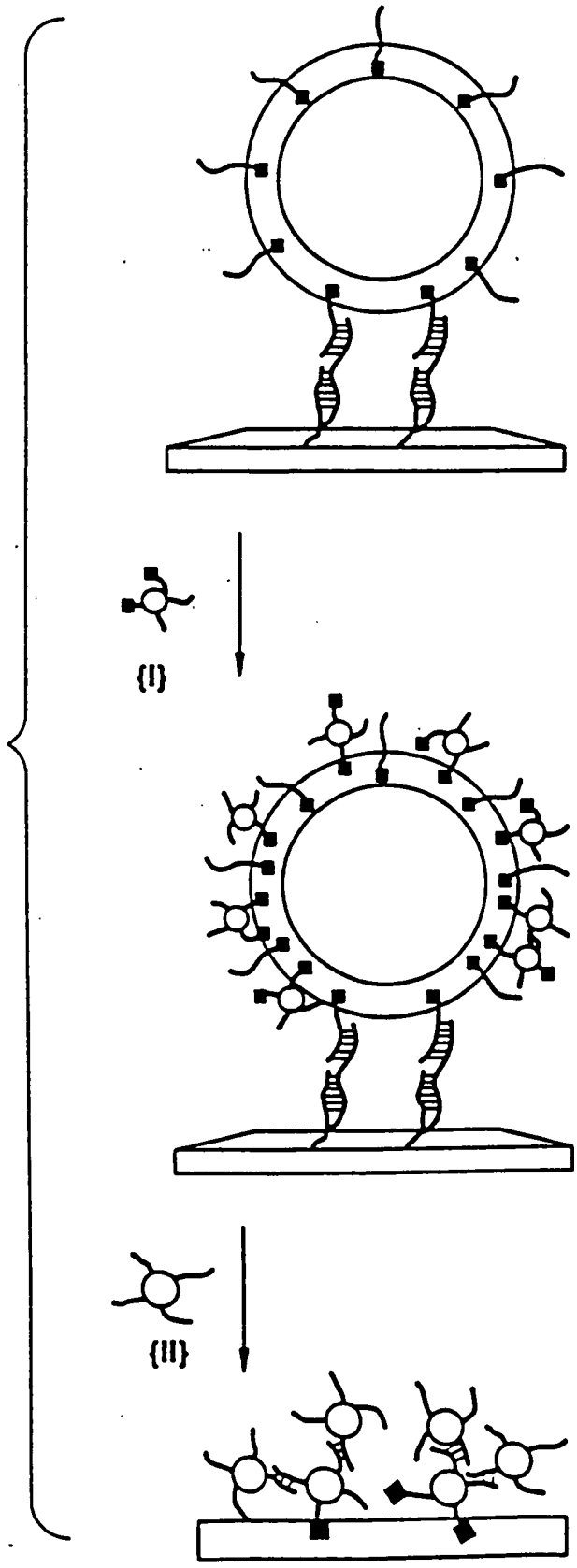


FIG.19A

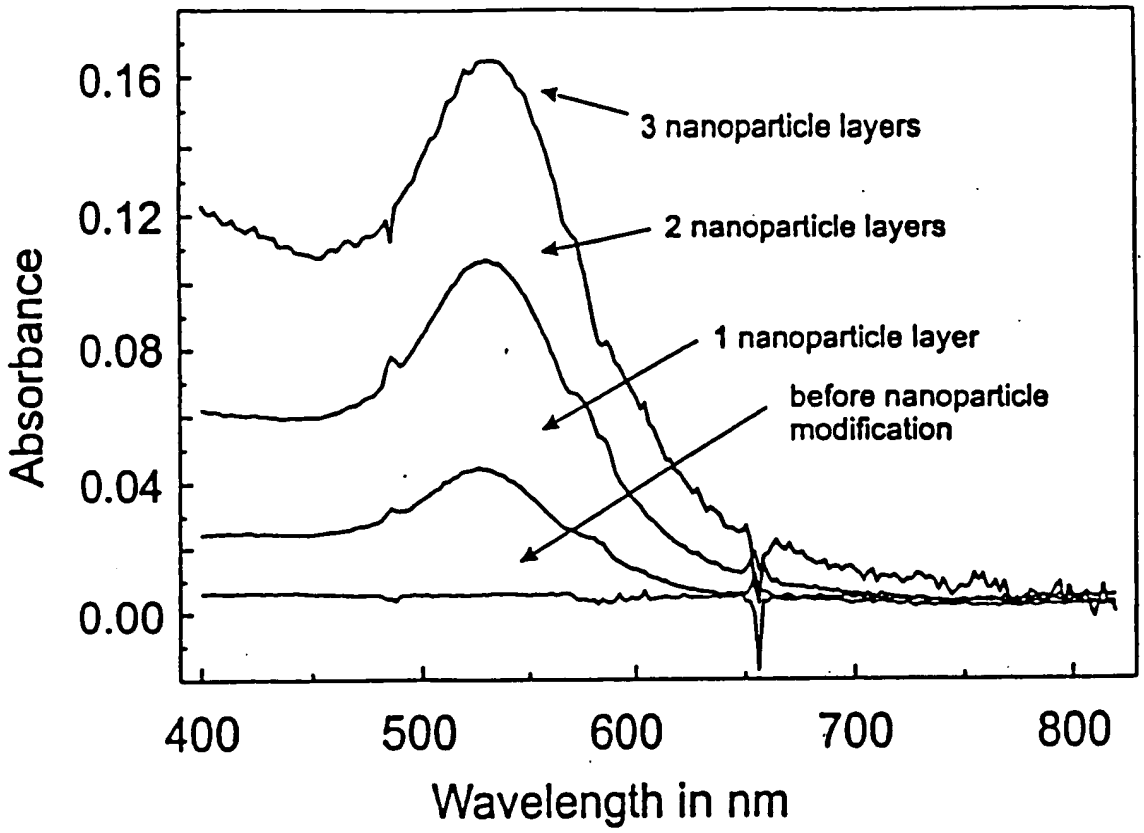


FIG.19B

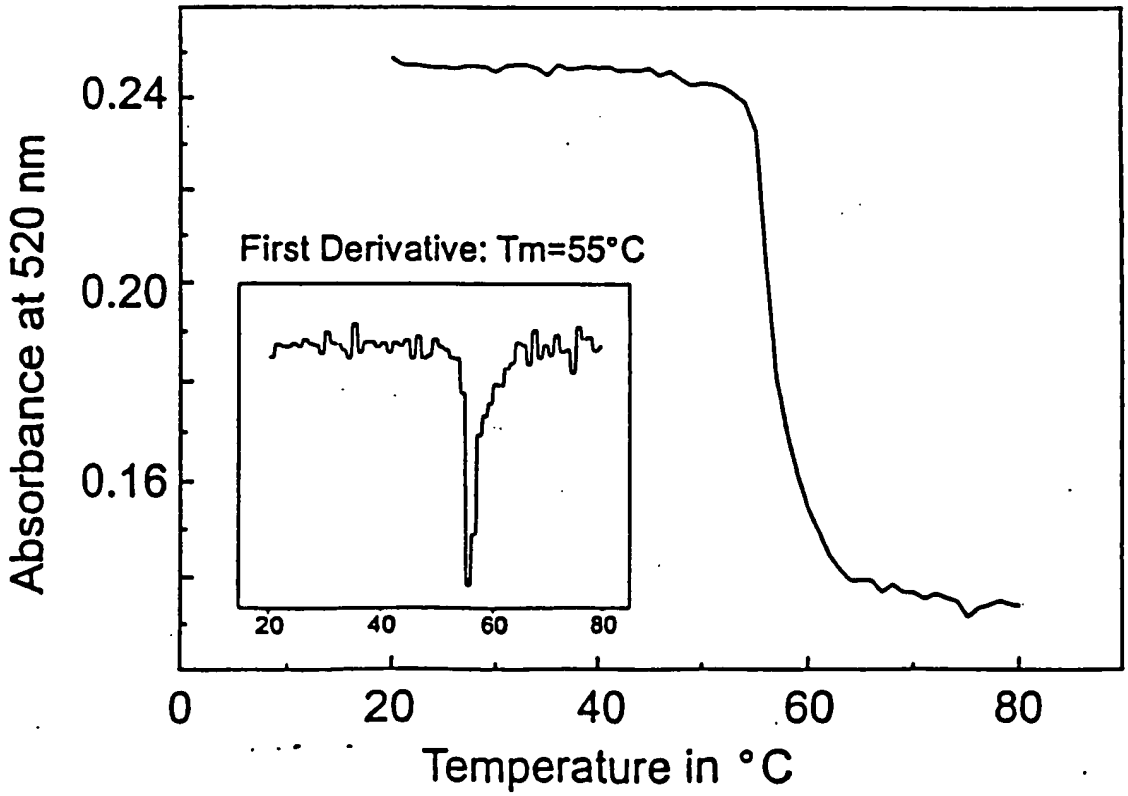


FIG.20A

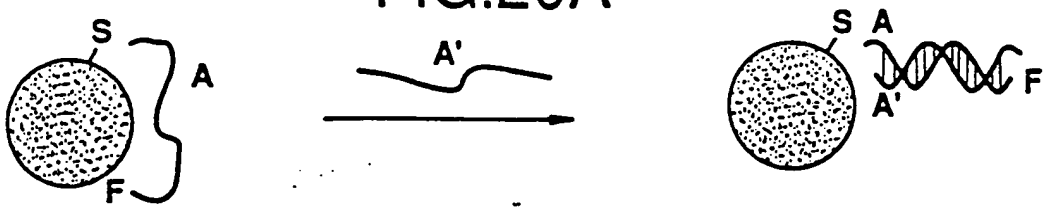
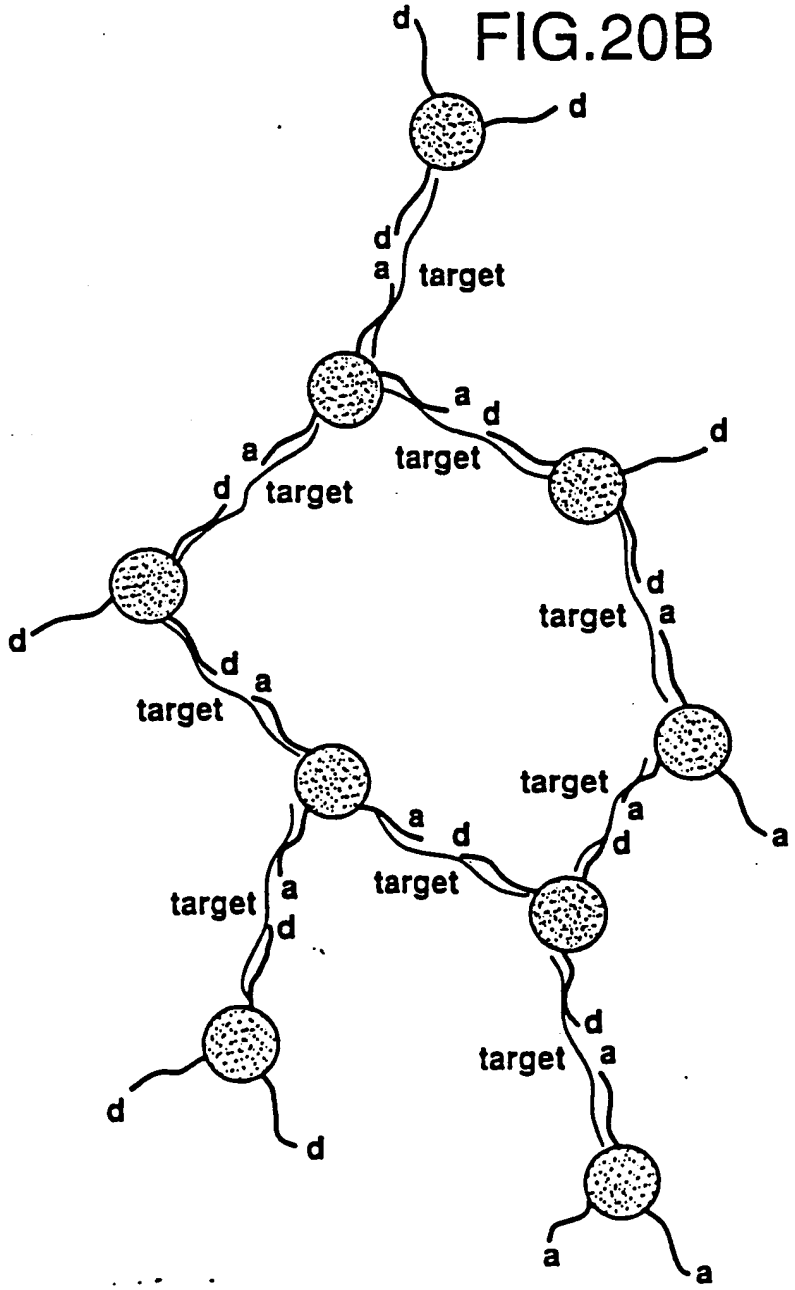


FIG.20B



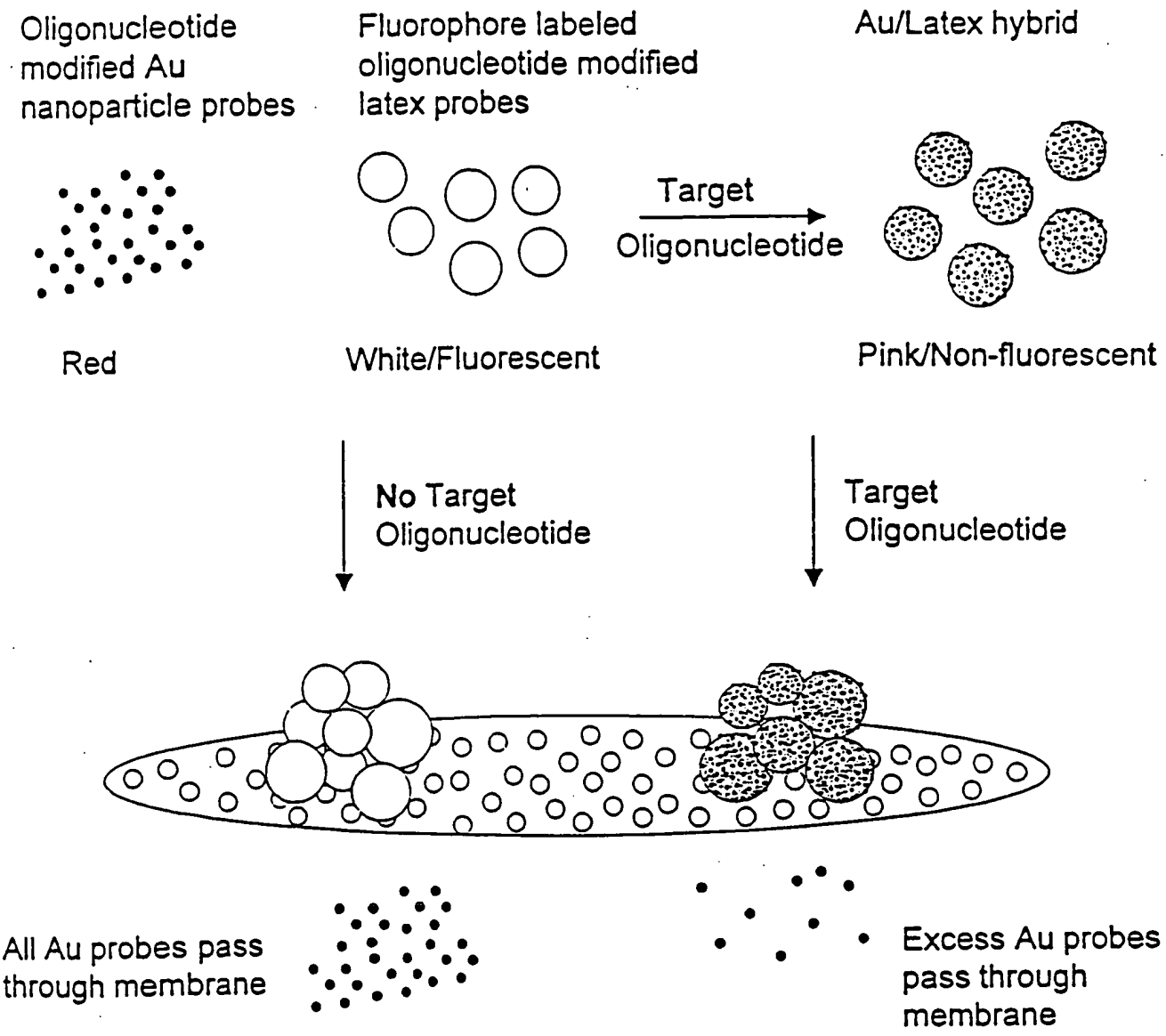
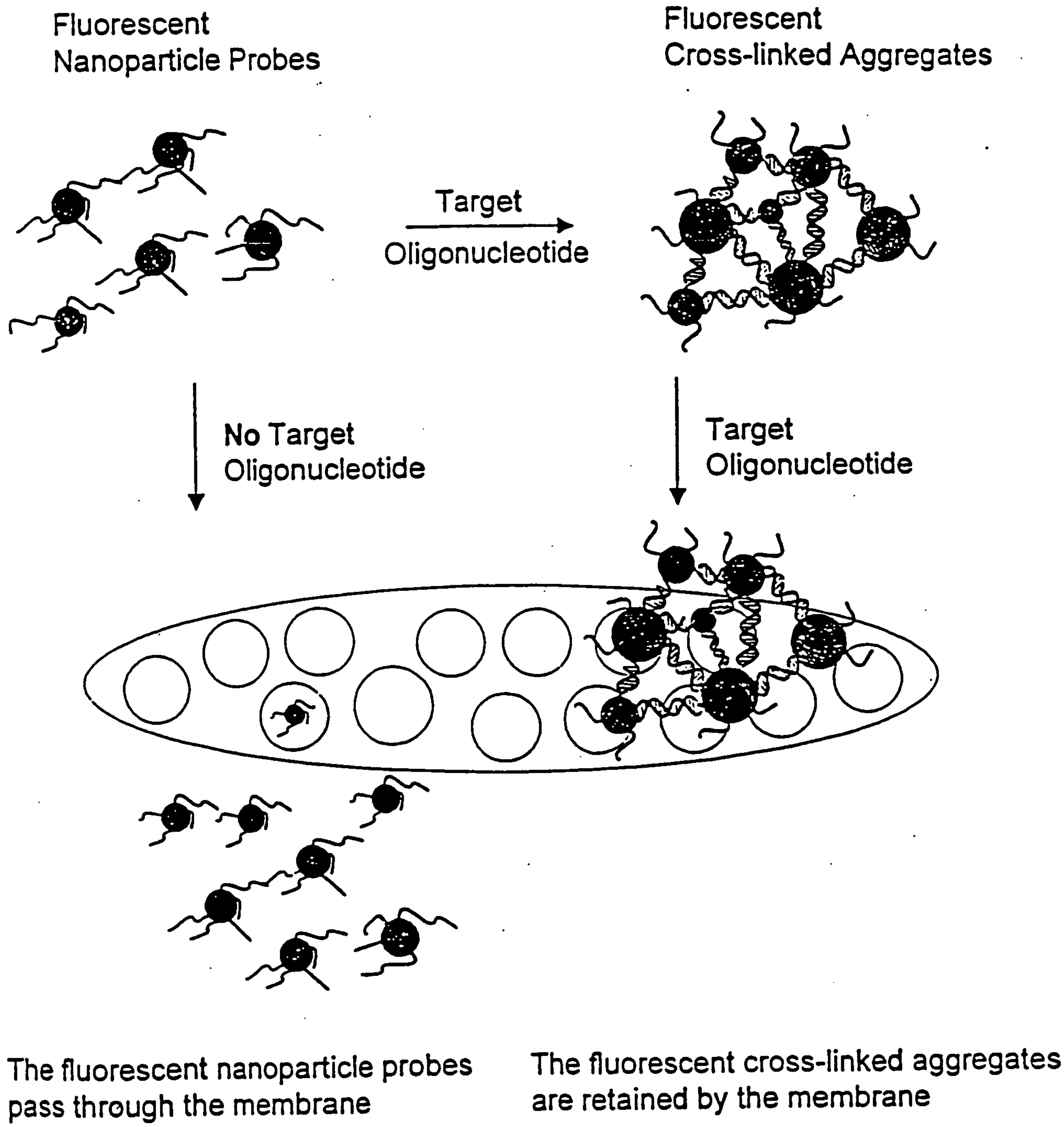

Oligonucleotide modified Au nanoparticle probes
Fluorophore labeled oligonucleotide modified latex probes
Au/Latex hybrid
Target Oligonucleotide
Red
White/Fluorescent
Pink/Non-fluorescent
No Target Oligonucleotide
Target Oligonucleotide
All Au probes pass through membrane
Excess Au probes pass through membrane

FIGURE 21

FIGURE 22


Fluorescent
Nanoparticle Probes
Fluorescent
Cross-linked Aggregates
Target
Oligonucleotide
No Target
Oligonucleotide
Target
Oligonucleotide
The fluorescent nanoparticle probes
pass through the membrane
The fluorescent cross-linked aggregates
are retained by the membrane

Anthrax PCR Product

5'G GCG GAT GAG TCA GTA GTT AAG GAG GCT CAT AGA GAA GTA ATT AAT
3'C CGC CTA CTC AGT CAT CAA TTC CTC CGA GTA TCT CTT CAT TAA TTA

TCG TCA ACA GAG GGA TTA TTG TTA AAT ATT GAT AAG GAT ATA AGA AAA
AGC AGT TGT CTC CCT AAT AAC AAT TTA TAA CTA TTC CTA TAT TCT TTT

ATA TTA TCC AGG GTT ATA TTG TAG AAA TTG AAG ATA CTG AAG GGC TT 3'
TAT AAT AGG TCC CAA TAT AAC ATC TTT AAC TTC TAT GAC TTC CCG AA 5'

141 mer Anthrax PCR product [SEQ ID NO:36]

3' CTC CCT AAT AAC AAT
[SEQ ID NO:37]

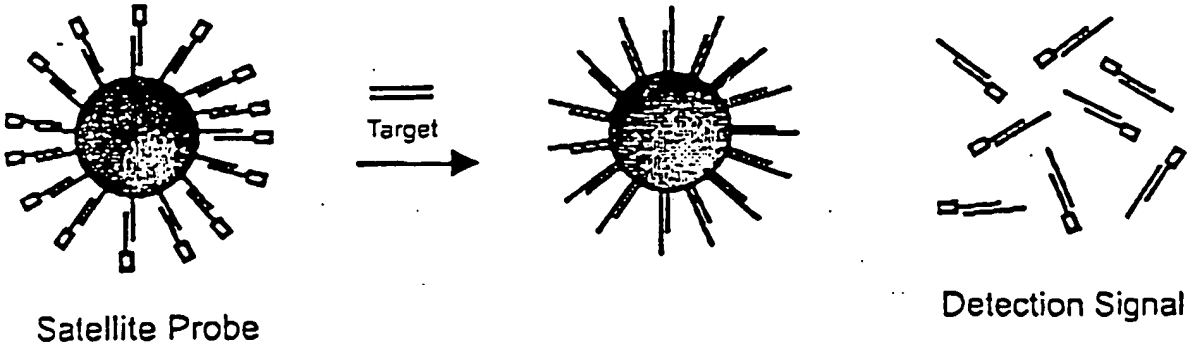
3' TTA TAA CTA TTC CTA
[SEQ ID NO:38]

Oligonucleotide-Nanoparticle Probes

Blocker Oligonucleotides

3' C CGC CTA CTC AGT CAT CAA TTC CTC CGA GT [SEQ ID NO:39]
3' A TCT CTT CAT TAA TTA AGC AGT TGT [SEQ ID NO:40]
3' TAT TCT TTT TAT AAT AGG TCC CAA TAT [SEQ ID NO:41]
3' AAC ATC TTT AAC TTC TAT GAC TTC CCG AA [SEQ ID NO:42]

FIGURE 23


Target
Satellite Probe
Detection Signal

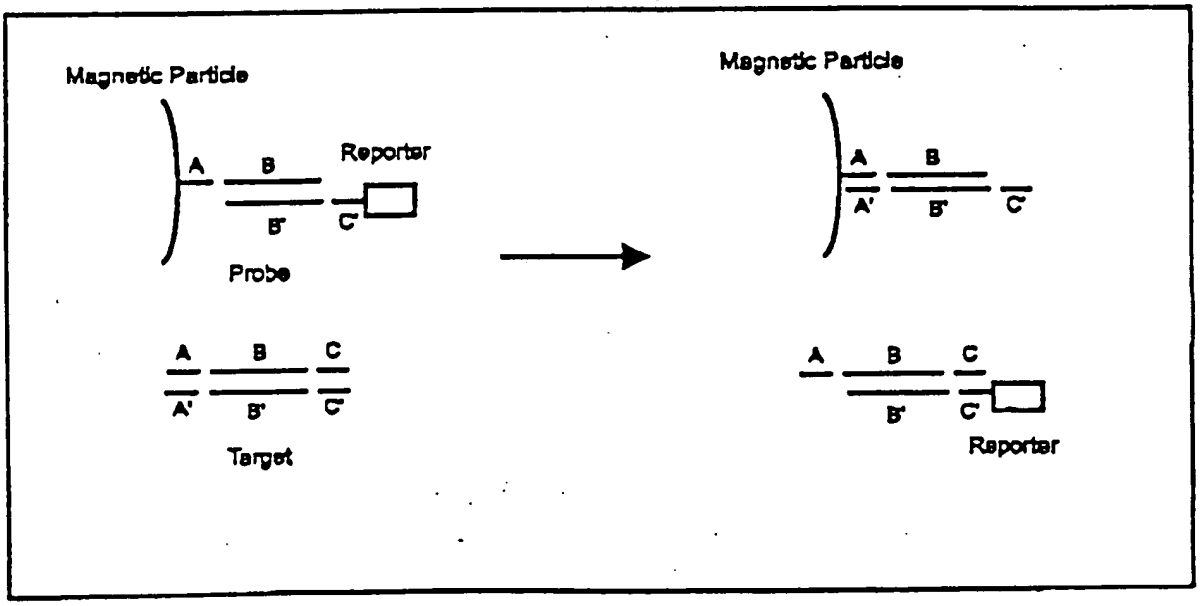

Magnetic Particle
A
B
Reporter
B'
C
Probe
A
B
C
A'
B'
C'
Target
Magnetic Particle
A
B
A'
B'
C'
A
B
C
B'
C'
Reporter

FIGURE 24

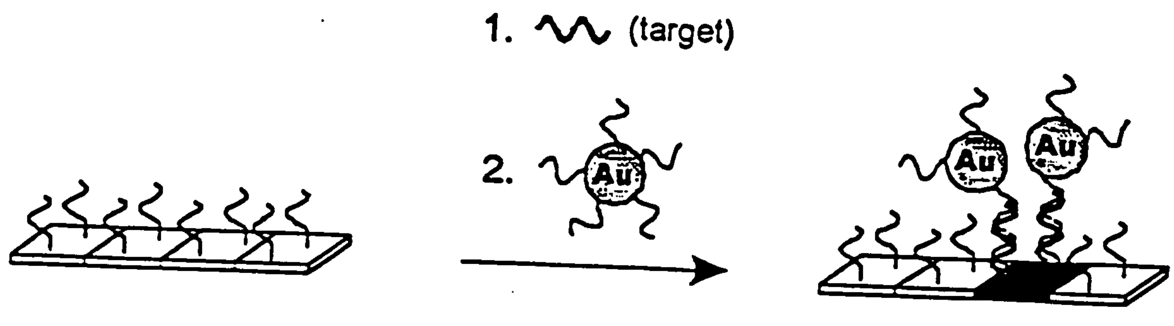


FIGURE 25A

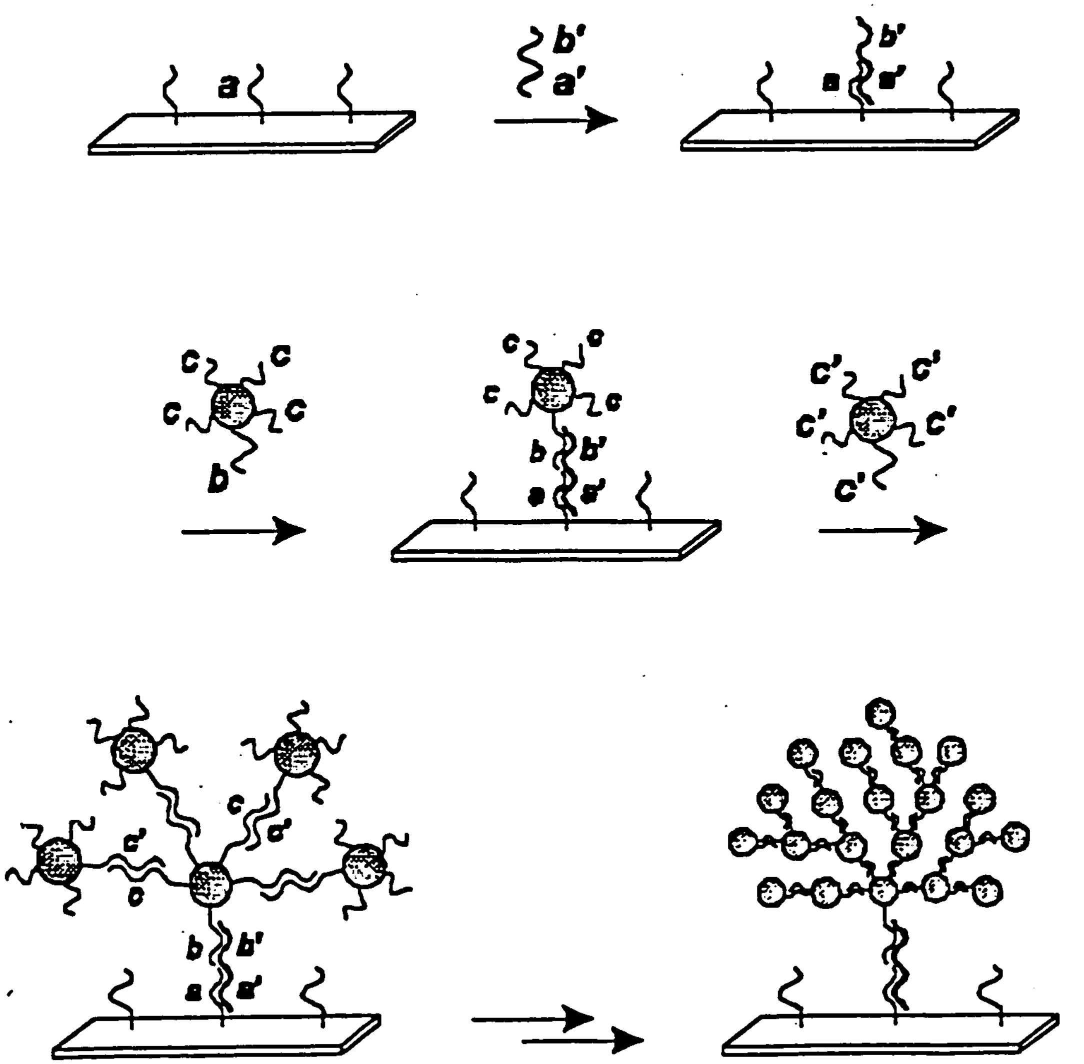
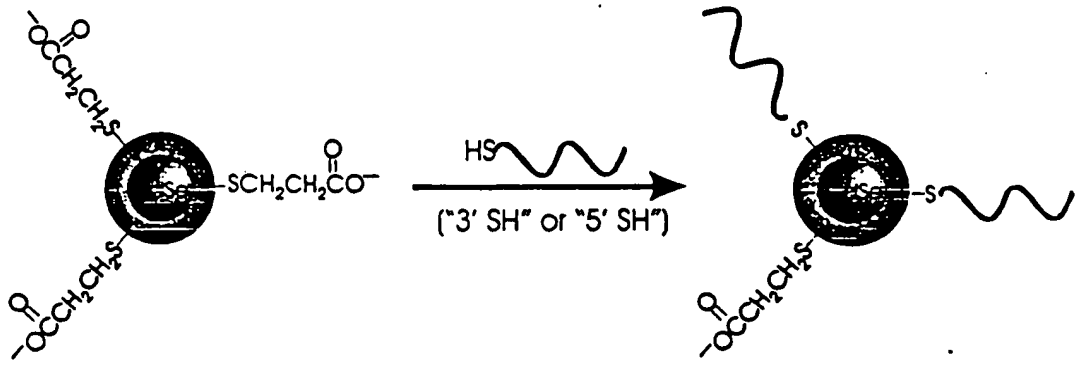
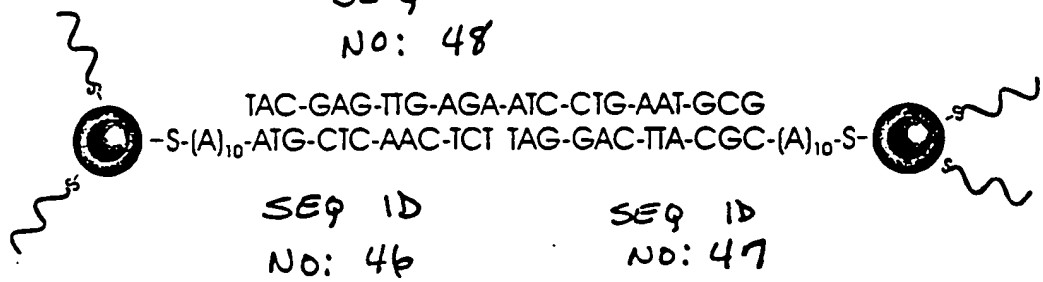


FIGURE 25D

A



B



SEQ ID NO: 48

SEQ ID NO: 46

SEQ ID NO: 47

FIGURE 26

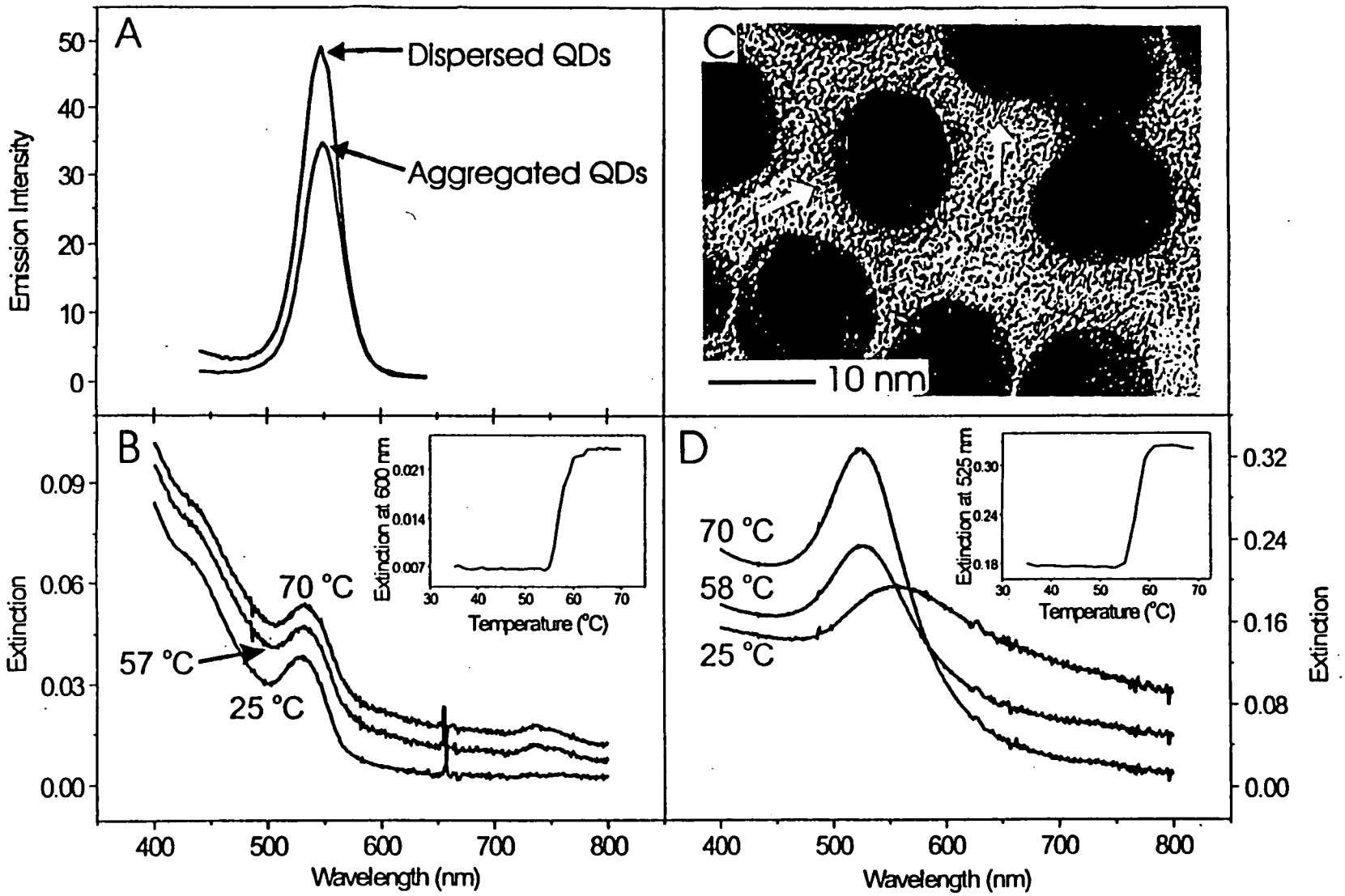

A
Dispersed QDs
Aggregated QDs
Emission Intensity
50
40
30
20
10
0
C
10 nm
B
70 °C
57 °C
25 °C
Extinction at 600 nm
0.021
0.014
0.007
30 40 50 60 70
Temperature (°C)
Extinction
0.09
0.06
0.03
0.00
400 500 600 700 800
Wavelength (nm)
D
70 °C
58 °C
25 °C
Extinction at 525 nm
0.30
0.24
0.18
30 40 50 60 70
Temperature (°C)
0.32
0.24
0.16
0.08
0.00
Extinction
400 500 600 700 800
Wavelength (nm)

FIGURE 27

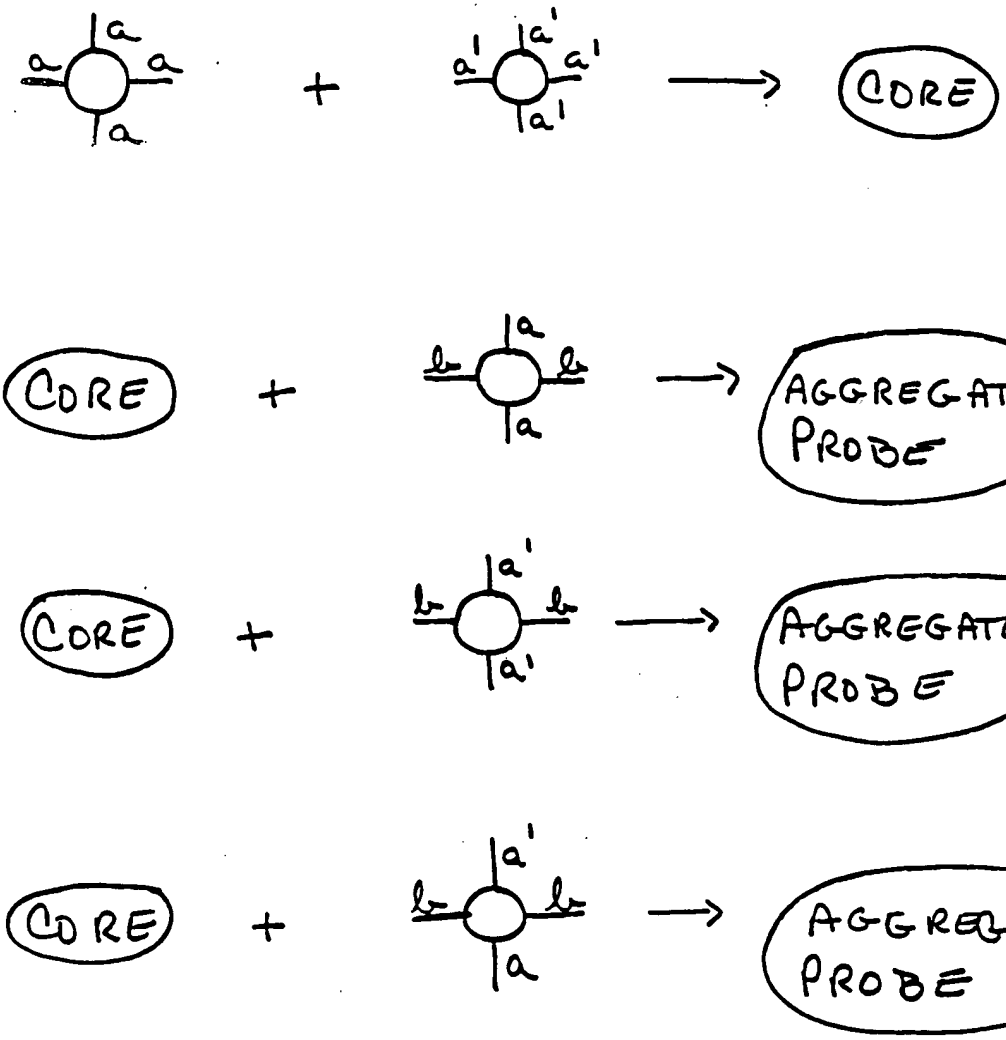


FIGURE 28A

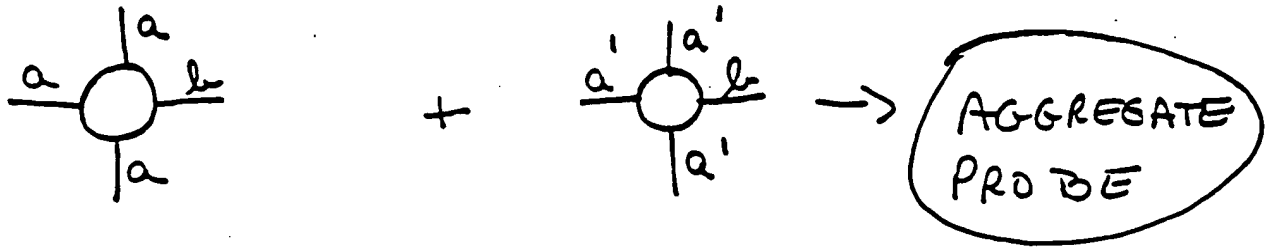


FIGURE 28 B

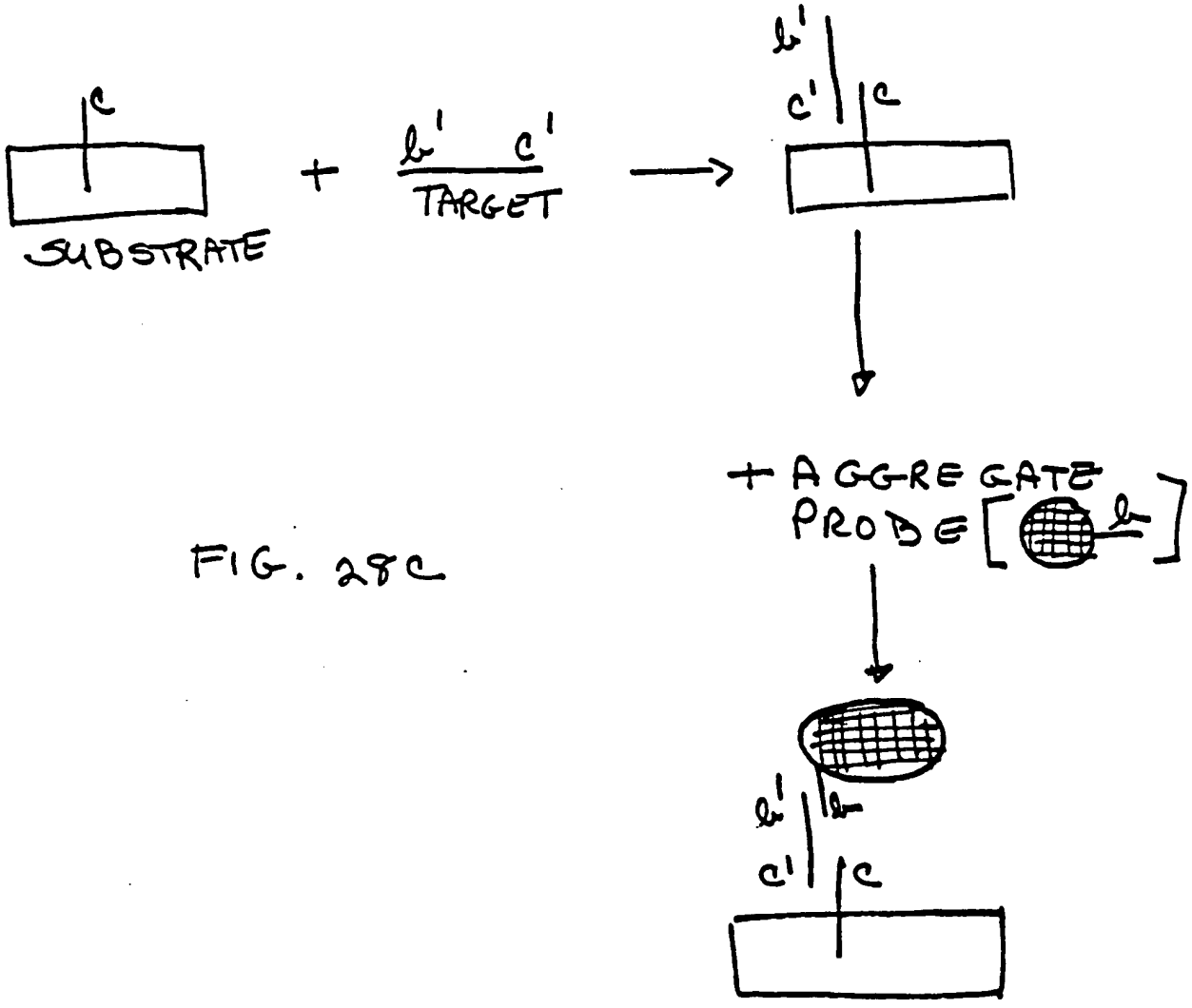


FIG. 28C

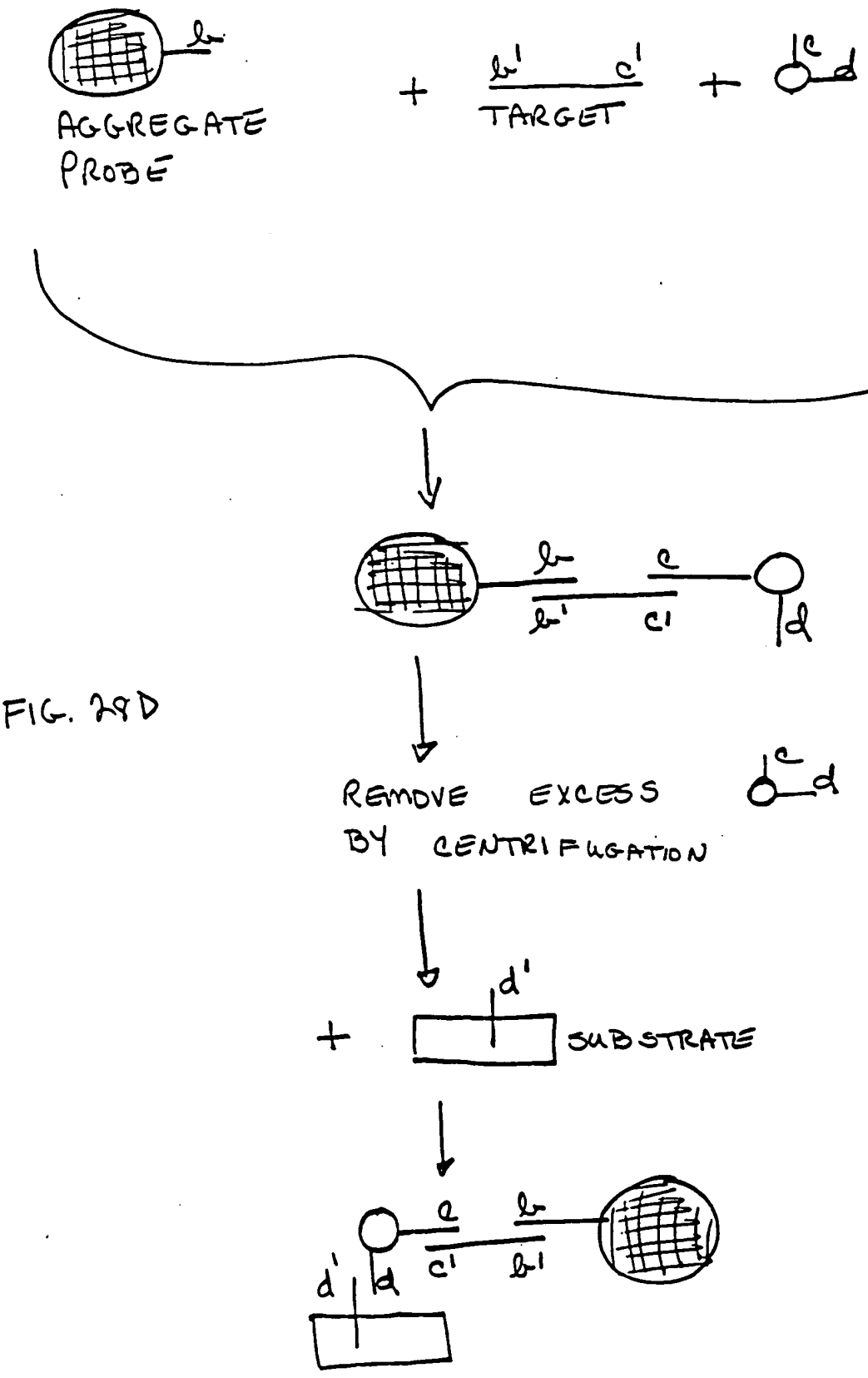

b
AGGREGATE
PROBE
+
b' c'
TARGET
+
c
d
b
c
b'
c'
d
REMOVE EXCESS
BY CENTRIFUGATION
c
d
d'
+
SUBSTRATE
c
b
c'
b'
d'
d

FIG. 28D

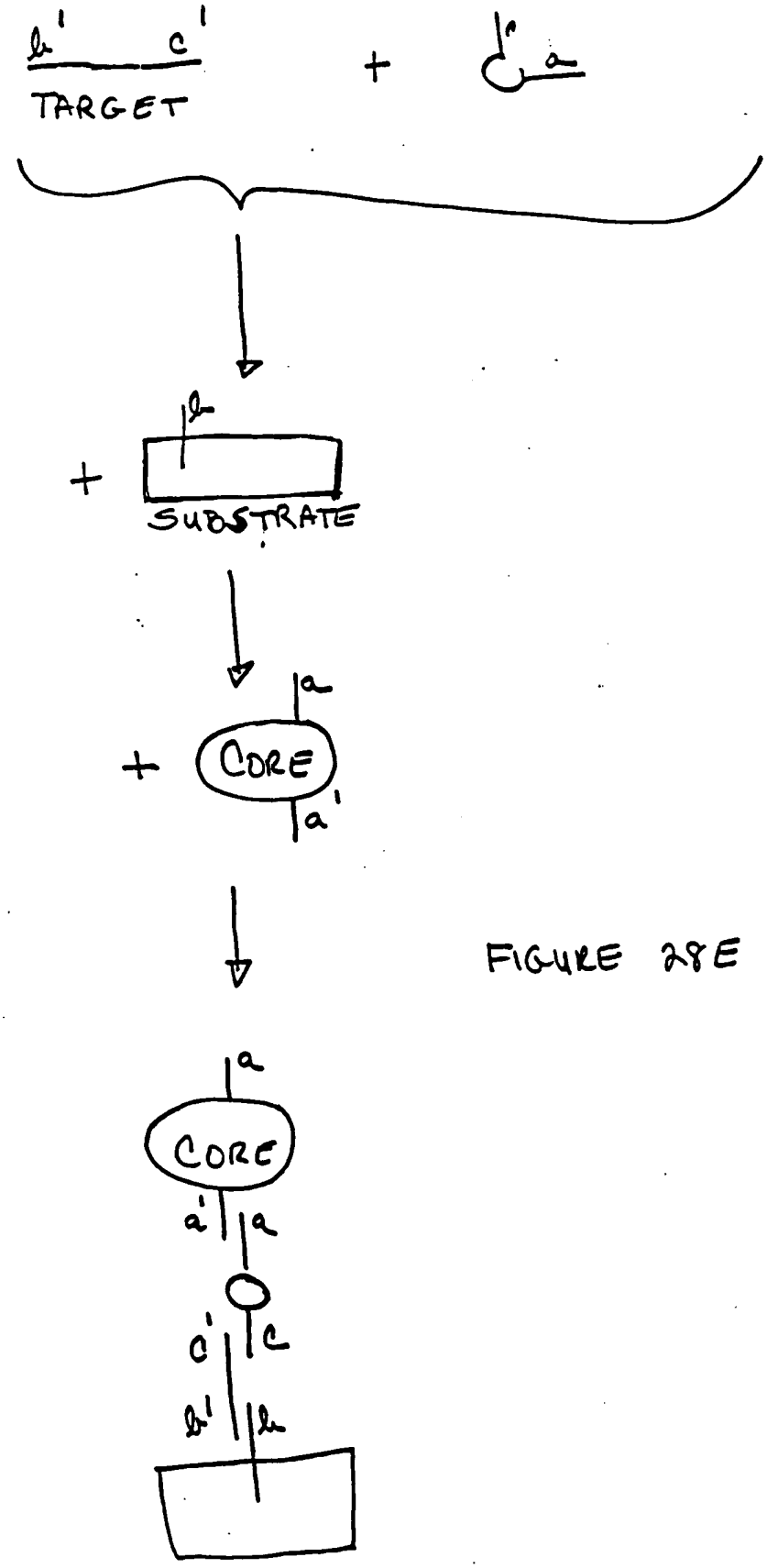


FIGURE 28E

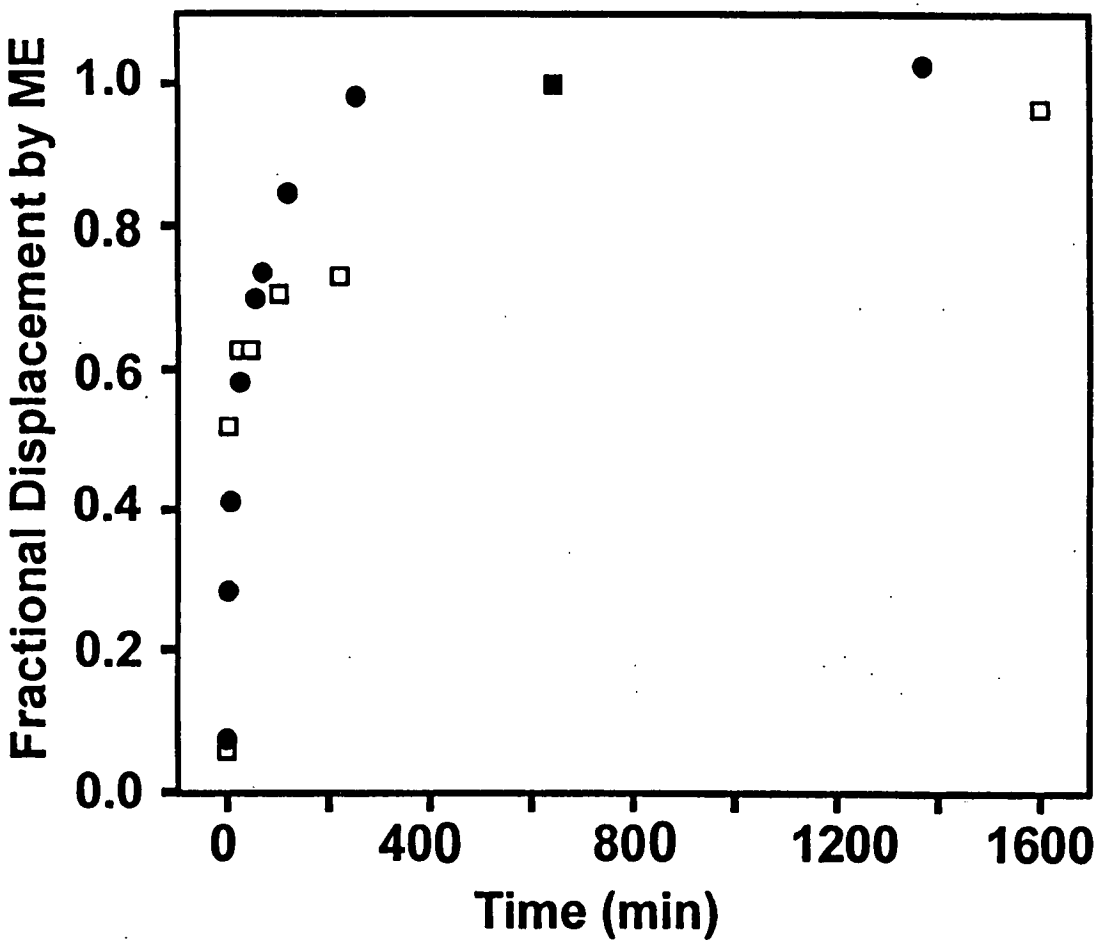

Fractional Displacement by ME
1.0
0.8
0.6
0.4
0.2
0.0
0
400
800
1200
1600
Time (min)

Figure 29

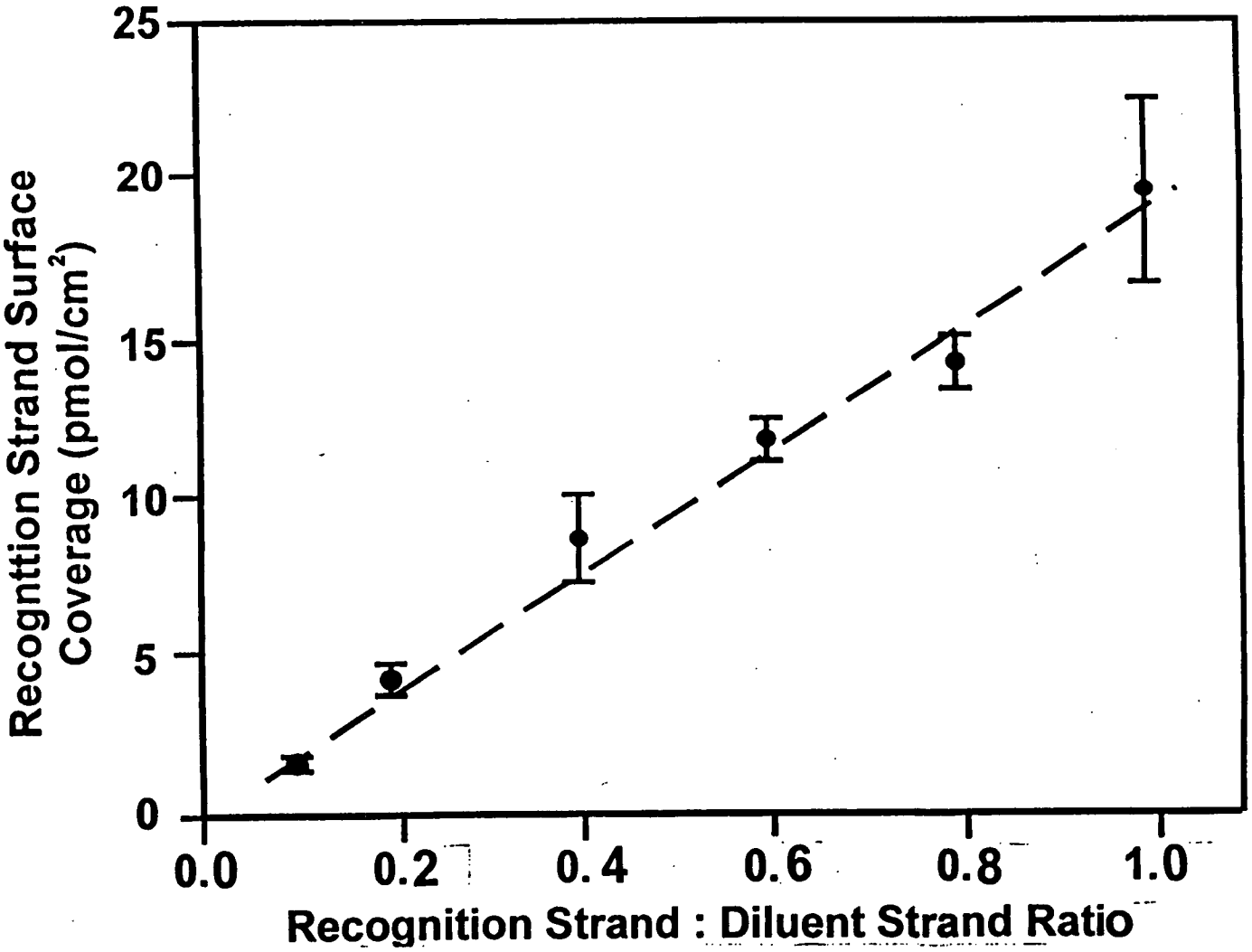

Recogntion Strand Surface Coverage (pmol/cm²)
25
20
15
10
5
0
0.0
0.2
0.4
0.6
0.8
1.0
Recognition Strand : Diluent Strand Ratio

Figure 30

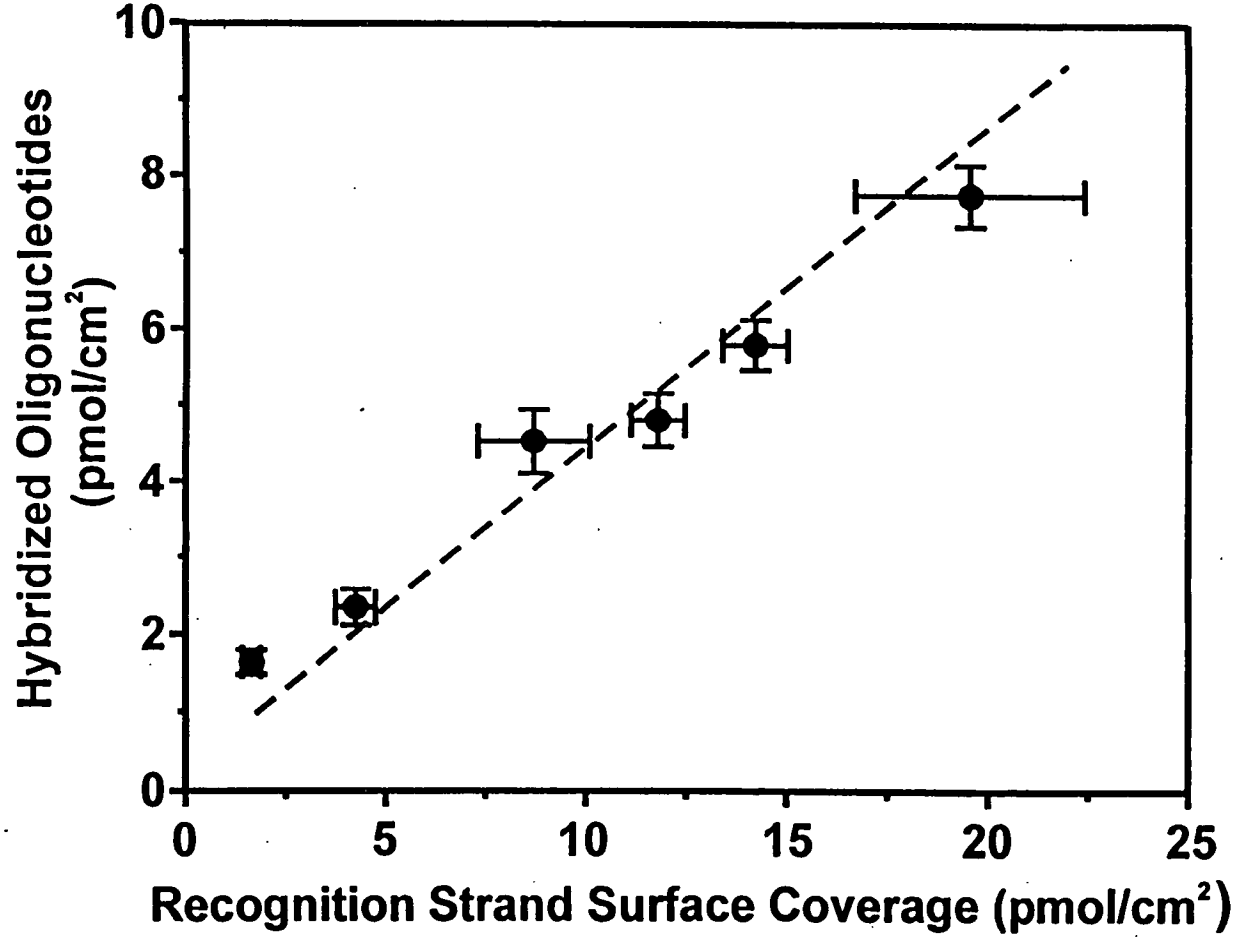
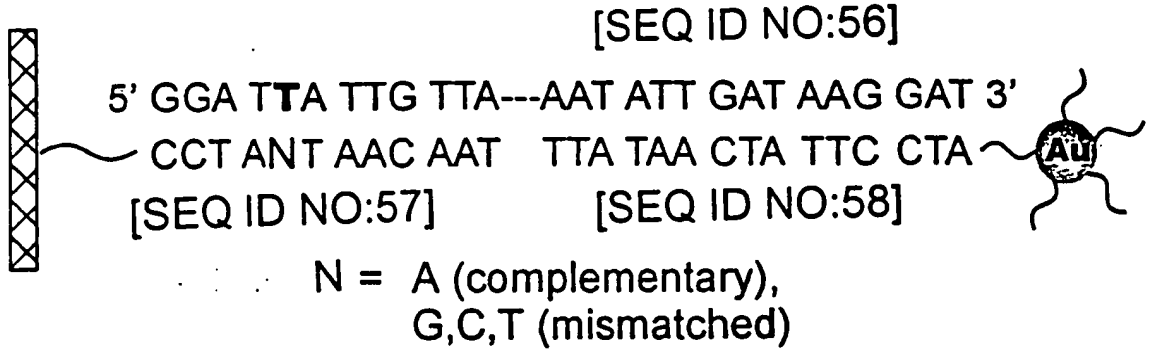

Hybridized Oligonucleotides (pmol/cm²)
0
2
4
6
8
10
0
5
10
15
20
25
Recognition Strand Surface Coverage (pmol/cm²)

Figure 31


[SEQ ID NO:56]
5' GGA TTA TTG TTA---AAT ATT GAT AAG GAT 3'
CCT ANT AAC AAT TTA TAA CTA TTC CTA
[SEQ ID NO:57]
[SEQ ID NO:58]
Au
N = A (complementary),
G,C,T (mismatched)

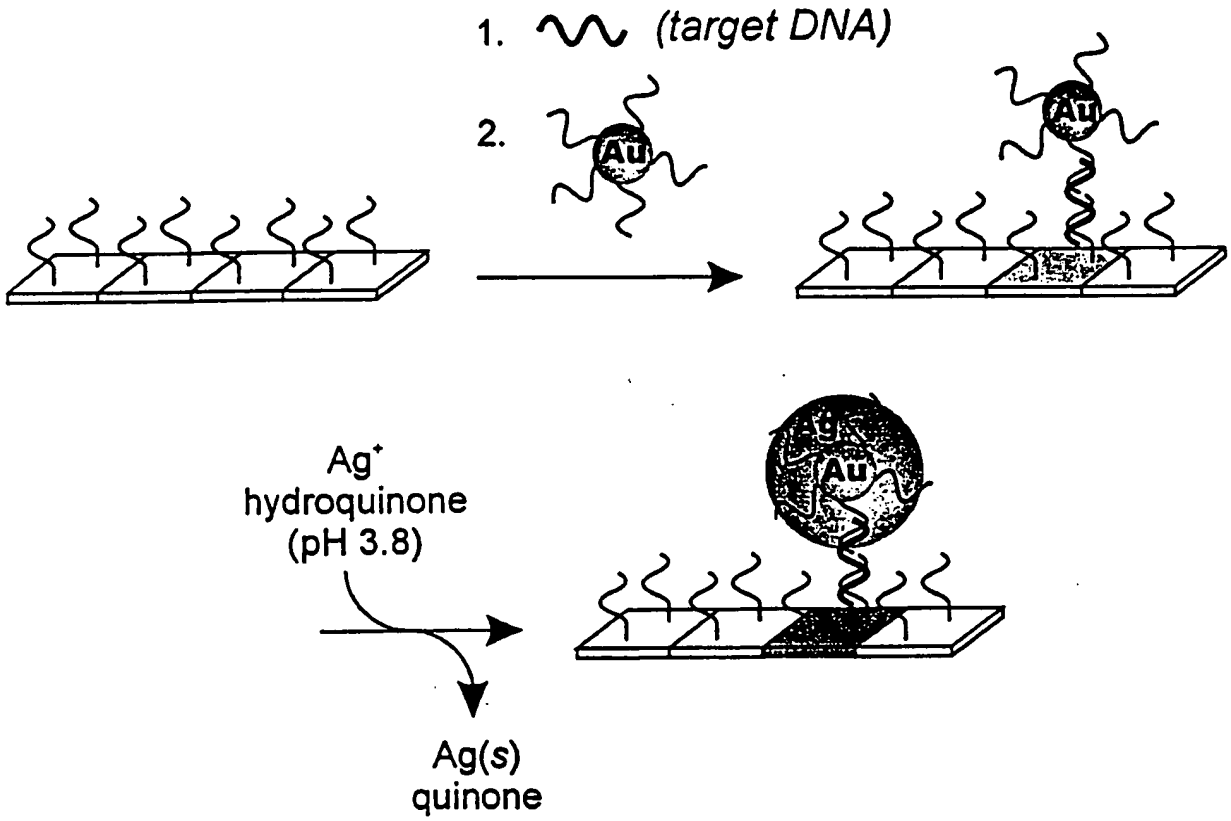

1. (target DNA)
2.
Au
Ag+
hydroquinone
(pH 3.8)
Ag(s)
quinone

Figure 32

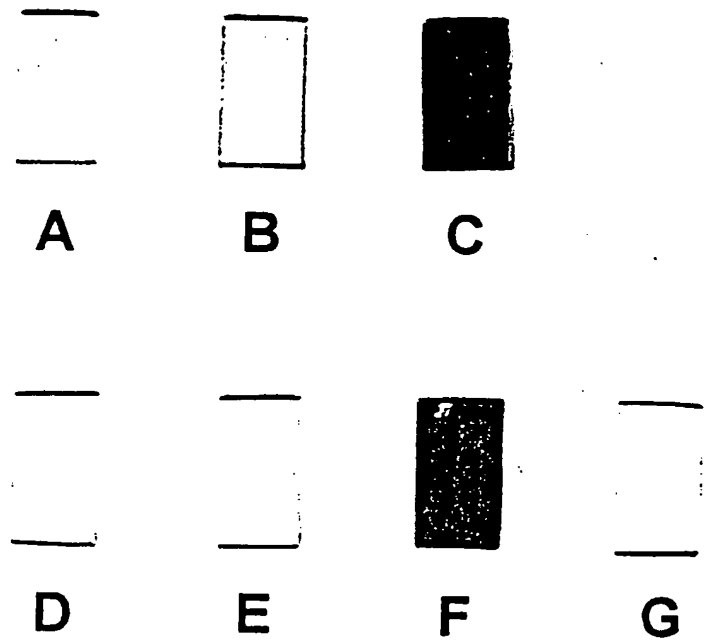


Figure 33

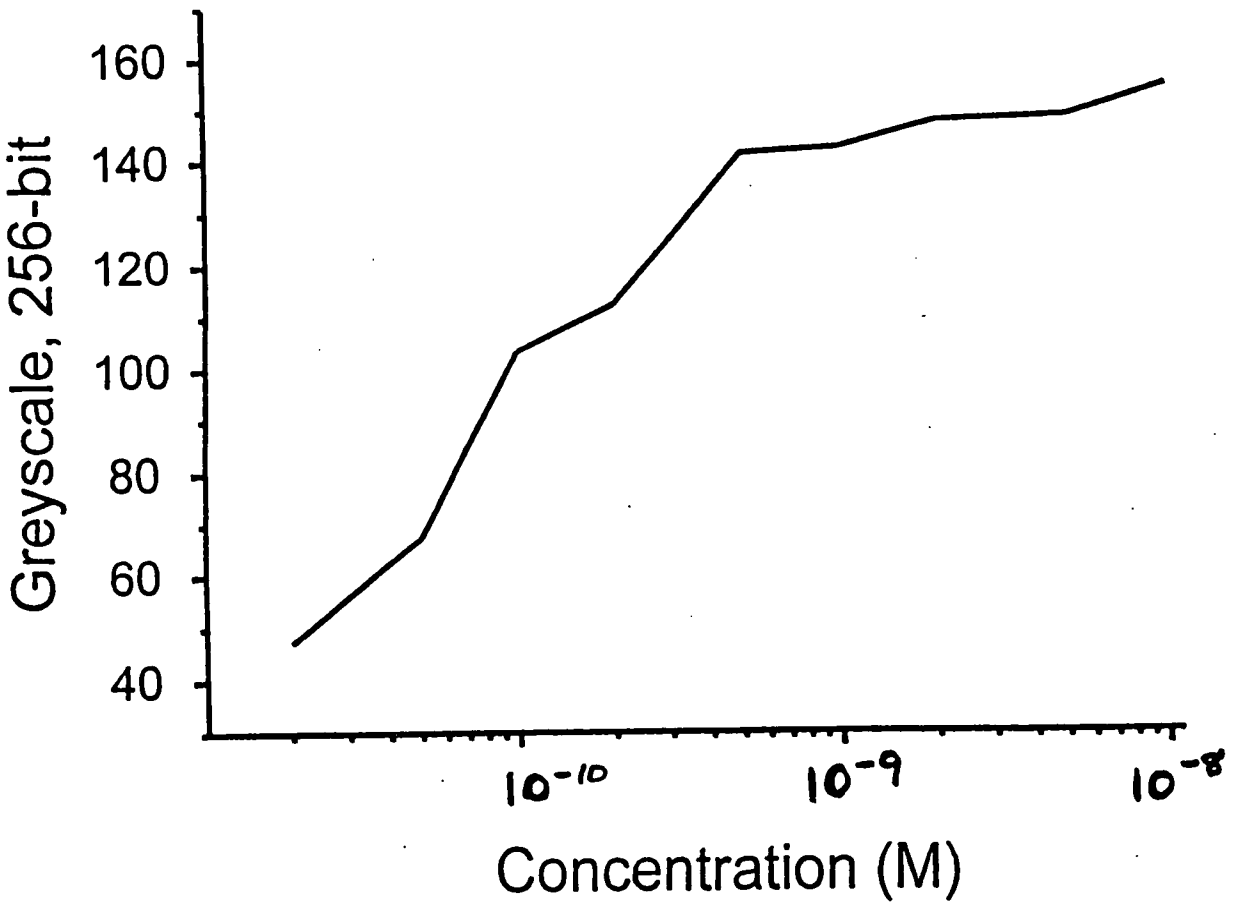

160
140
120
100
80
60
40
Greyscale, 256-bit
10-10
10-9
10-8
Concentration (M)

Figure 34

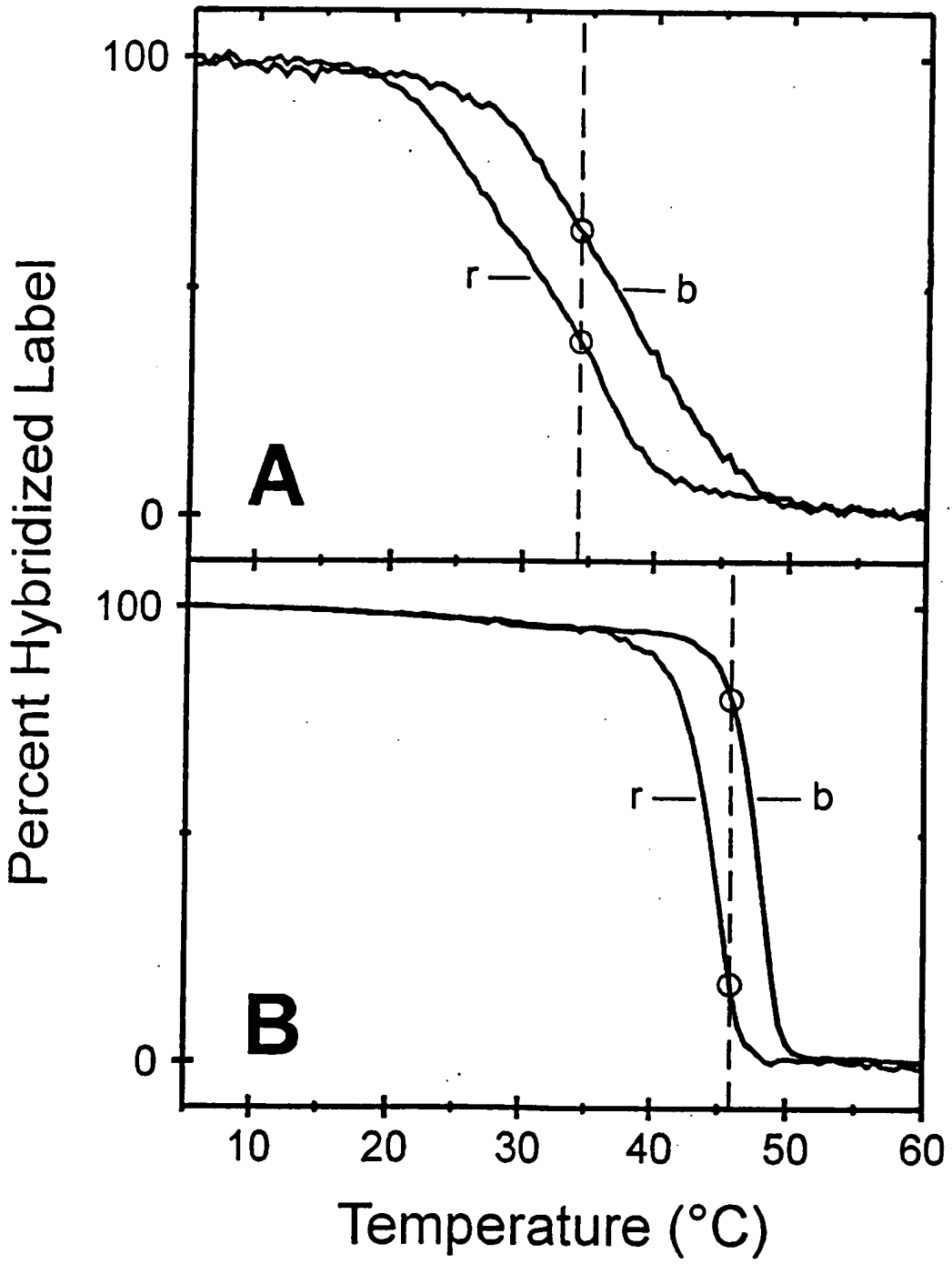

100
0
r
b
A
100
0
r
b
B
10
20
30
40
50
60
Percent Hybridized Label
Temperature (°C)

Figure 35

FIG. 36A

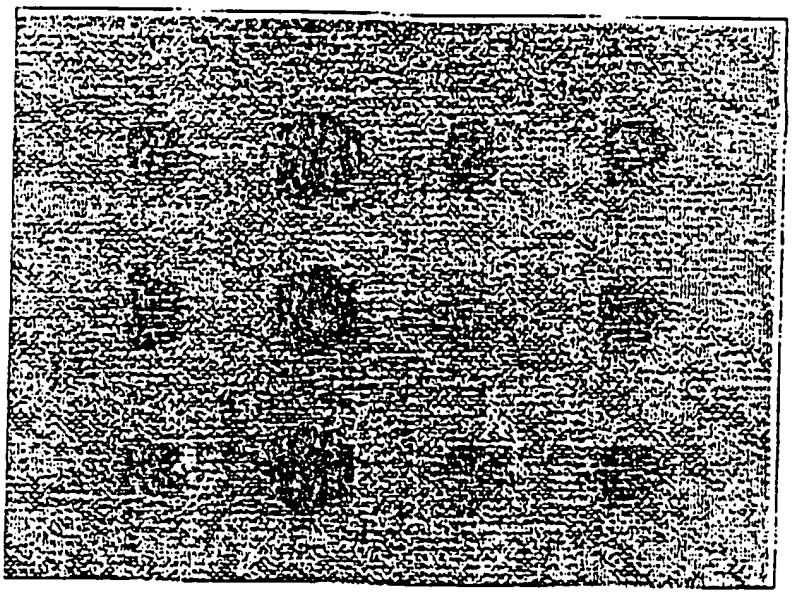
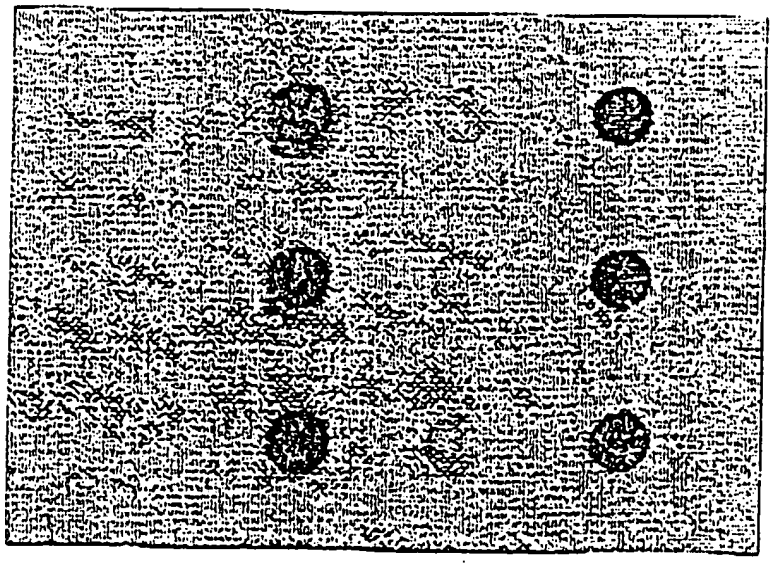


FIG. 36B



C A T G

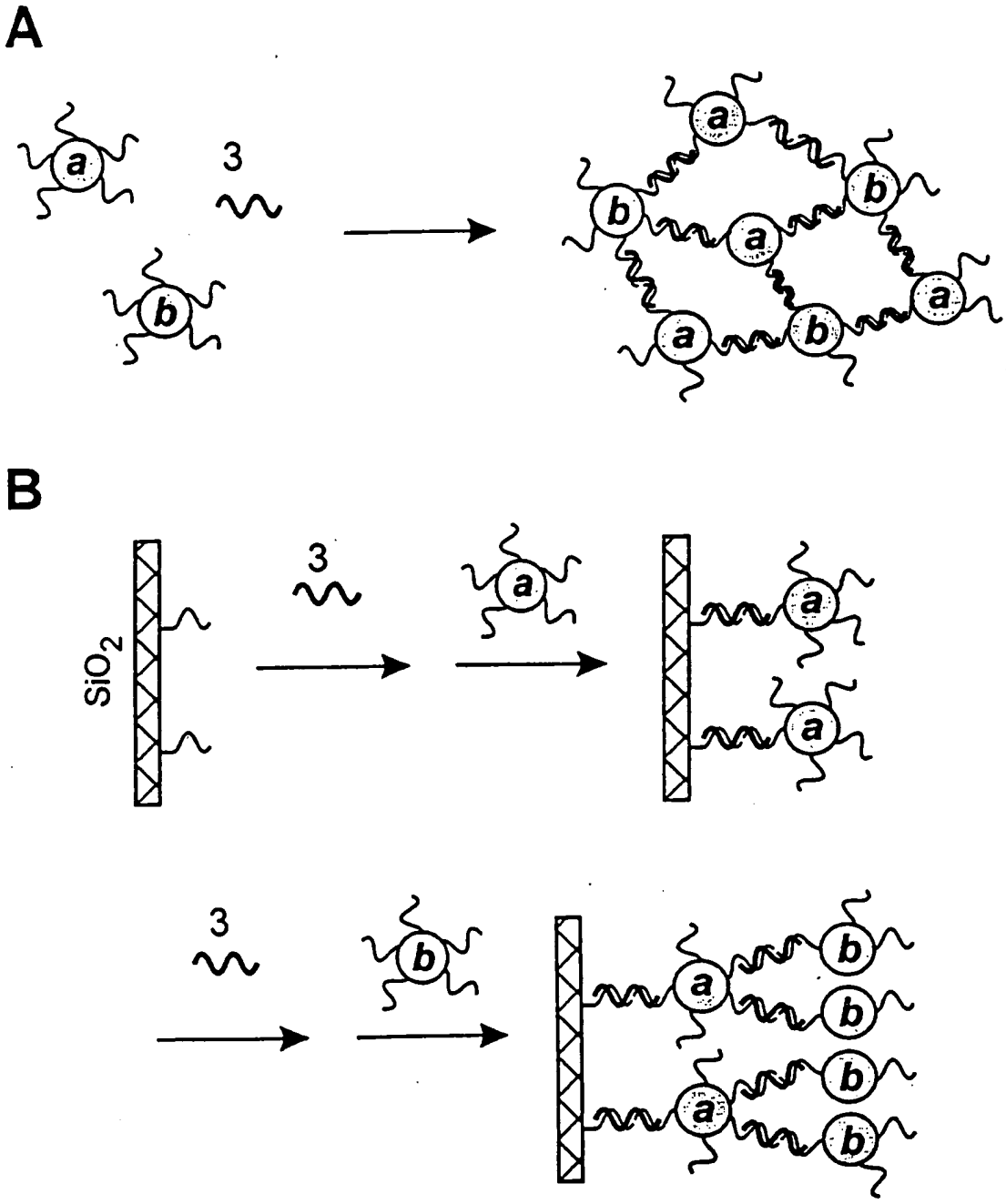


Figure 37

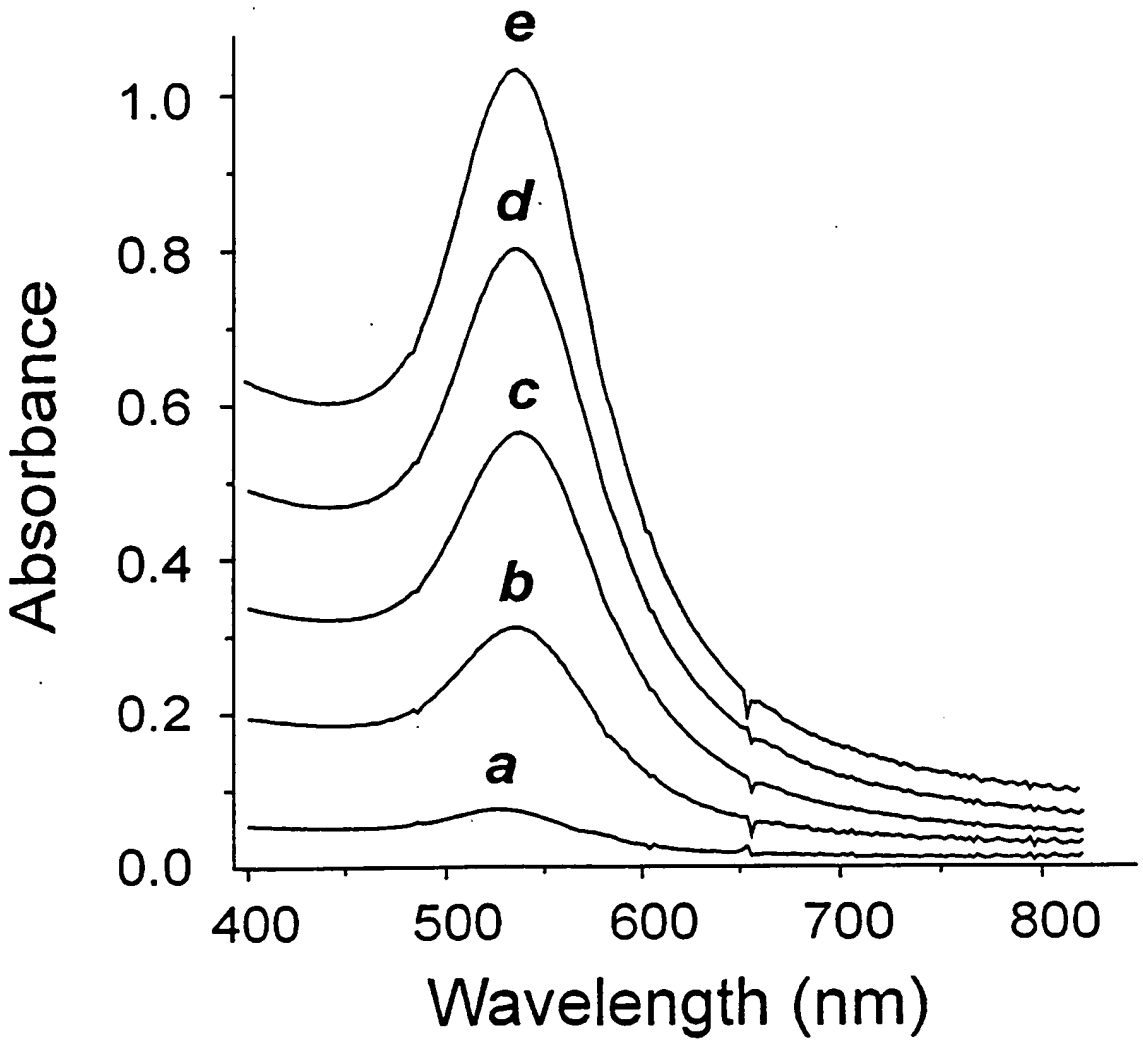

e
d
c
b
a
1.0
0.8
0.6
0.4
0.2
0.0
Absorbance
400
500
600
700
800
Wavelength (nm)

Figure 38A

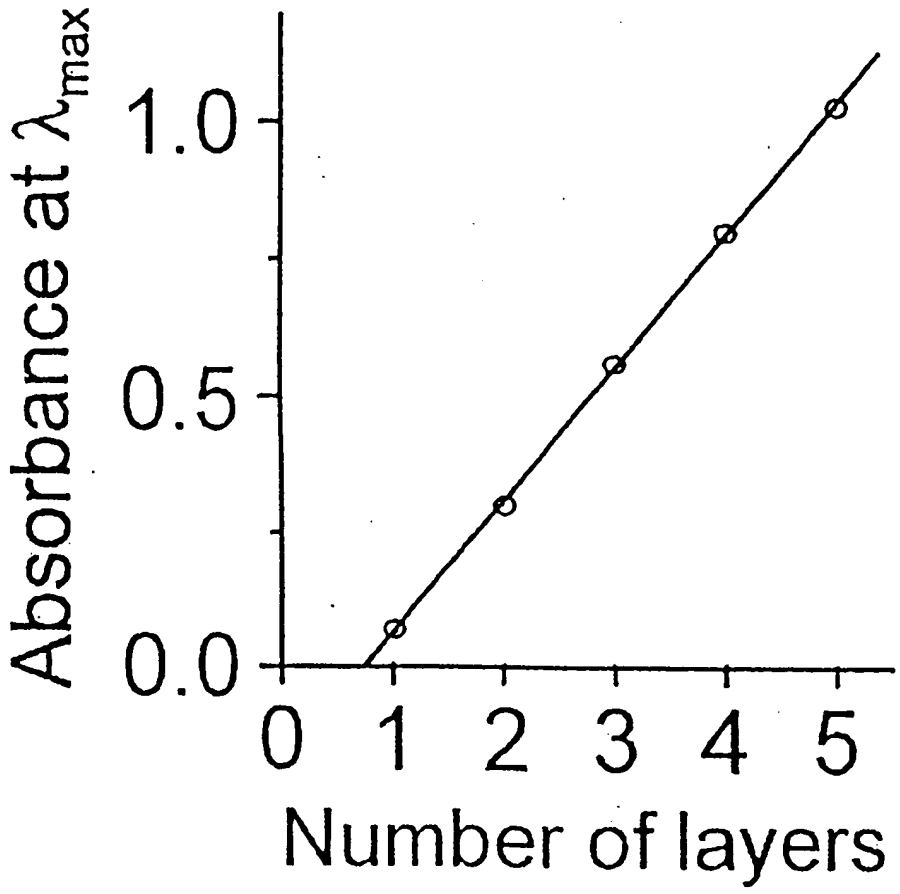


Figure 38B

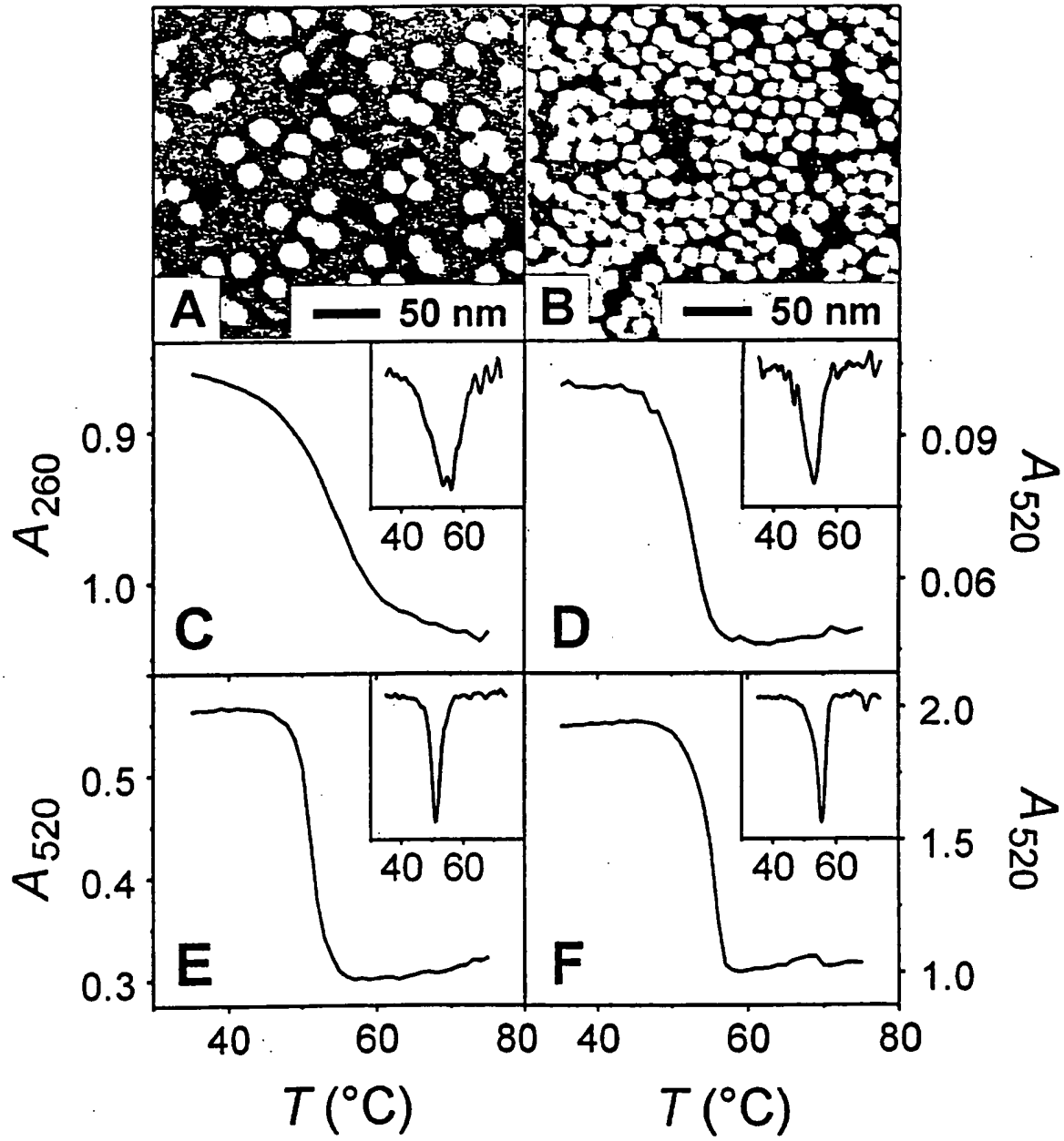

A
50 nm
B
50 nm
C
D
E
F
A_{260}
A_{520}
A_{520}
A_{520}
0.9
1.0
0.09
0.06
0.5
0.4
0.3
2.0
1.5
1.0
40 60
40 60
40 60
40 60
40
60
80
40
60
80
T (°C)
T (°C)

Figure 39

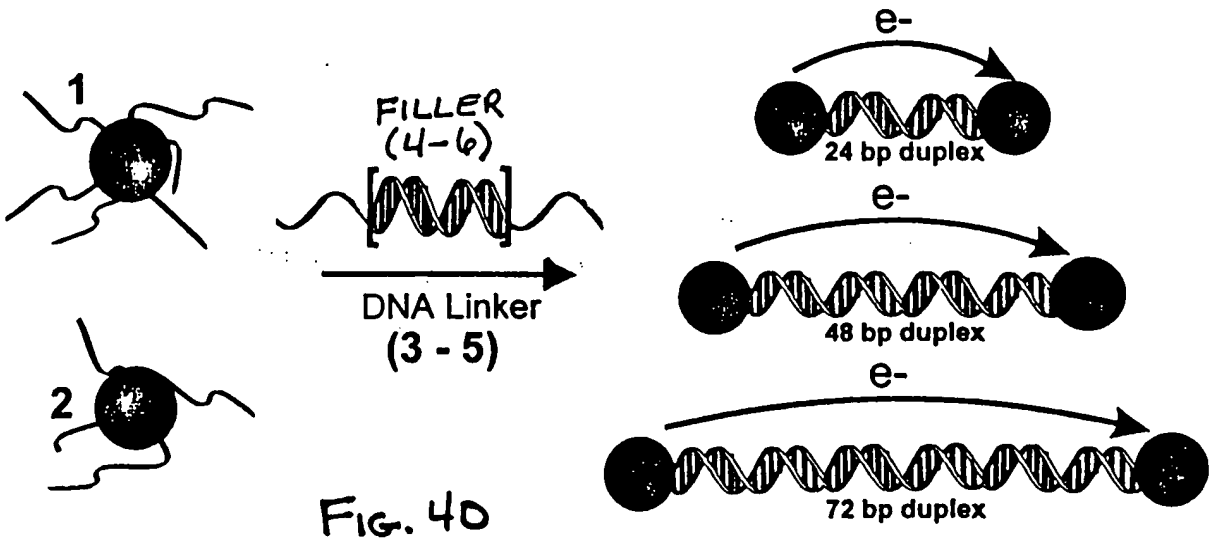

1
2
FILLER
(4-6)
DNA Linker
(3 - 5)
e-
24 bp duplex
e-
48 bp duplex
e-
72 bp duplex

FIG. 40

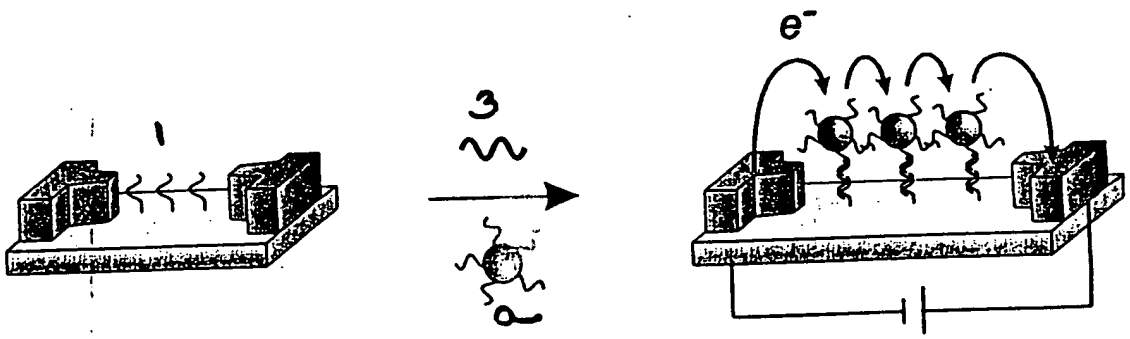

1
3
e-

FIG. 41